



US007915484B2

(12) **United States Patent**  
**da Costa e Silva et al.**

(10) **Patent No.:** **US 7,915,484 B2**  
(45) **Date of Patent:** **Mar. 29, 2011**

(54) **PROTEIN KINASE STRESS-RELATED PROTEINS AND METHODS OF USE IN PLANTS**

(75) Inventors: **Oswaldo da Costa e Silva**, Neustadt (DE); **Nocha Van Thielen**, Cary, NC (US); **Ruoying Chen**, Duluth, GA (US); **Hans J. Bohnert**, Champaign, IL (US); **Rodrigo Sarria-Millan**, West Lafayette, IN (US)

(73) Assignee: **BASF Plant Science GmbH** (DE)

(\* ) Notice: Subject to any disclaimer, the term of this patent is extended or adjusted under 35 U.S.C. 154(b) by 0 days.

(21) Appl. No.: **12/873,345**

(22) Filed: **Sep. 1, 2010**

(65) **Prior Publication Data**

US 2010/0325759 A1 Dec. 23, 2010

**Related U.S. Application Data**

(60) Division of application No. 09/828,313, filed on Apr. 6, 2001, now Pat. No. 6,867,351, and a continuation of application No. 12/545,903, filed on Aug. 24, 2009, now Pat. No. 7,858,847, and a continuation of application No. 12/401,635, filed on Mar. 11, 2009, and a continuation of application No. 11/961,634, filed on Dec. 20, 2007, now Pat. No. 7,521,598, and a continuation of application No. 11/564,902, filed on Nov. 30, 2006, now Pat. No. 7,504,559, and a continuation of application No. 10/768,863, filed on Jan. 30, 2004, now Pat. No. 7,179,962.

(60) Provisional application No. 60/196,001, filed on Apr. 7, 2000.

(51) **Int. Cl.**  
**C12N 15/82** (2006.01)  
**C12N 15/29** (2006.01)  
**A01H 5/00** (2006.01)  
**A01H 5/10** (2006.01)

(52) **U.S. Cl.** ..... **800/289**; 435/419; 536/23.6; 800/298; 800/320; 800/320.1; 800/320.2; 800/320.3; 800/312; 800/314; 800/306; 800/322; 800/313; 800/317; 800/317.1; 800/317.2; 800/317.3; 800/317.4

(58) **Field of Classification Search** ..... None  
See application file for complete search history.

(56) **References Cited**

U.S. PATENT DOCUMENTS

7,084,323 B1 \* 8/2006 Sheen ..... 800/289

\* cited by examiner

*Primary Examiner* — Cynthia Collins

(74) *Attorney, Agent, or Firm* — Patricia A. McDaniels

(57) **ABSTRACT**

A transgenic plant transformed by a Protein Kinase Stress-Related Protein (PKSRP) coding nucleic acid, wherein expression of the nucleic acid sequence in the plant results in increased tolerance to environmental stress as compared to a wild type variety of the plant. Also provided are agricultural products, including seeds, produced by the transgenic plants. Also provided are isolated PKSRPs, and isolated nucleic acid coding PKSRPs, and vectors and host cells containing the latter.

**22 Claims, 18 Drawing Sheets**

Figure 1

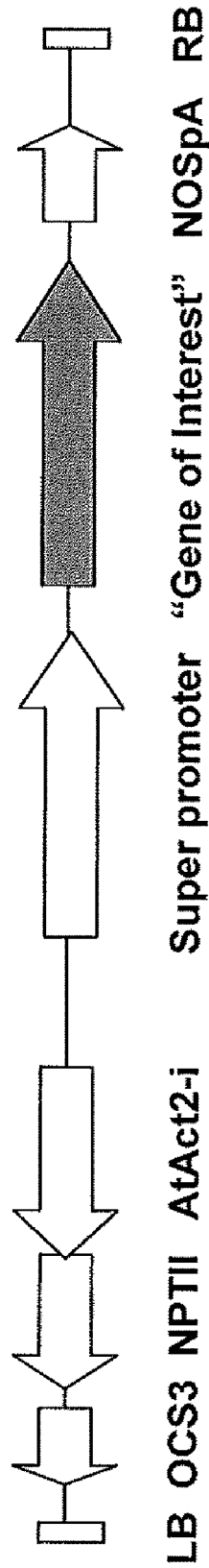


Figure 2

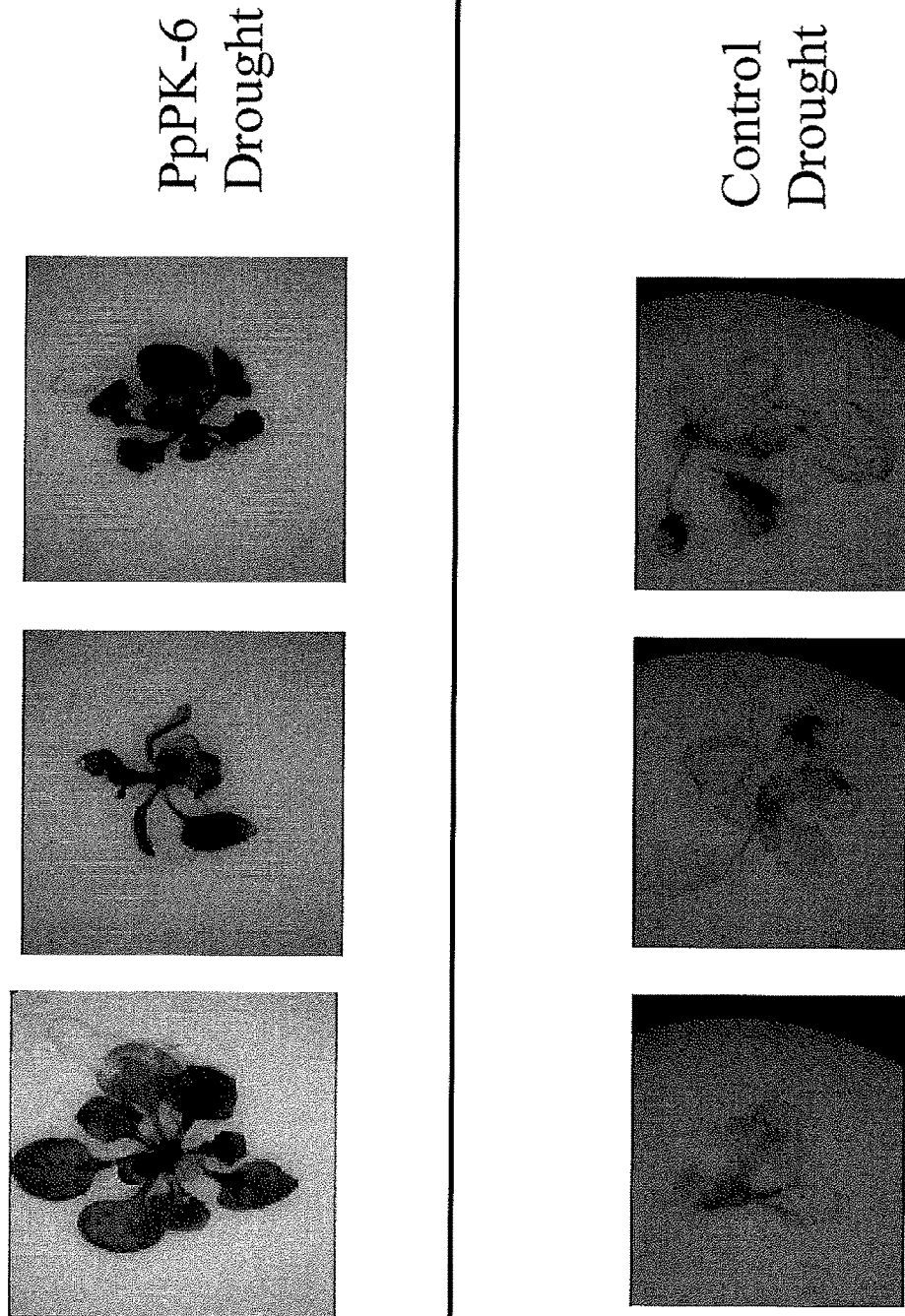


Figure 3

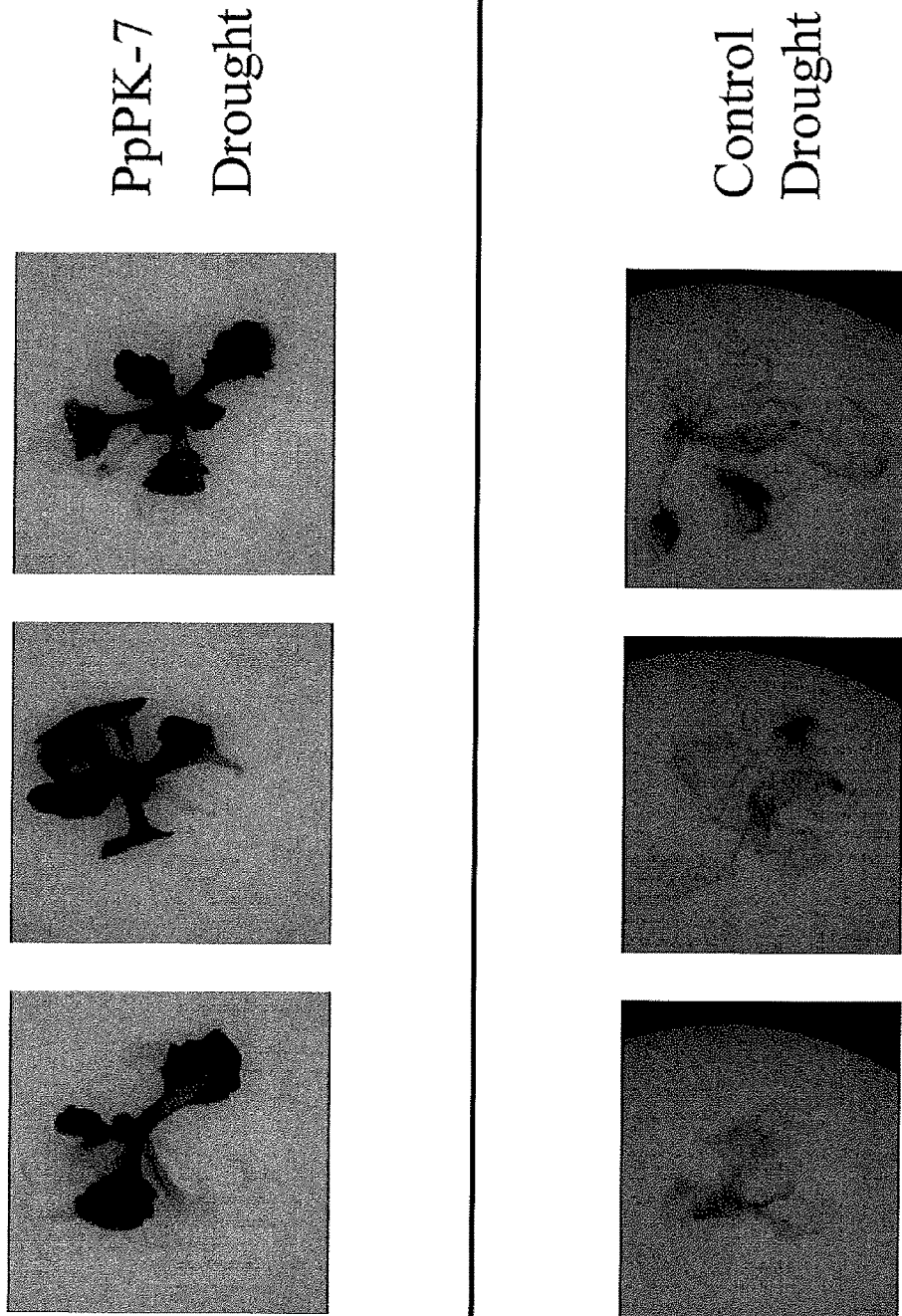


Figure 4

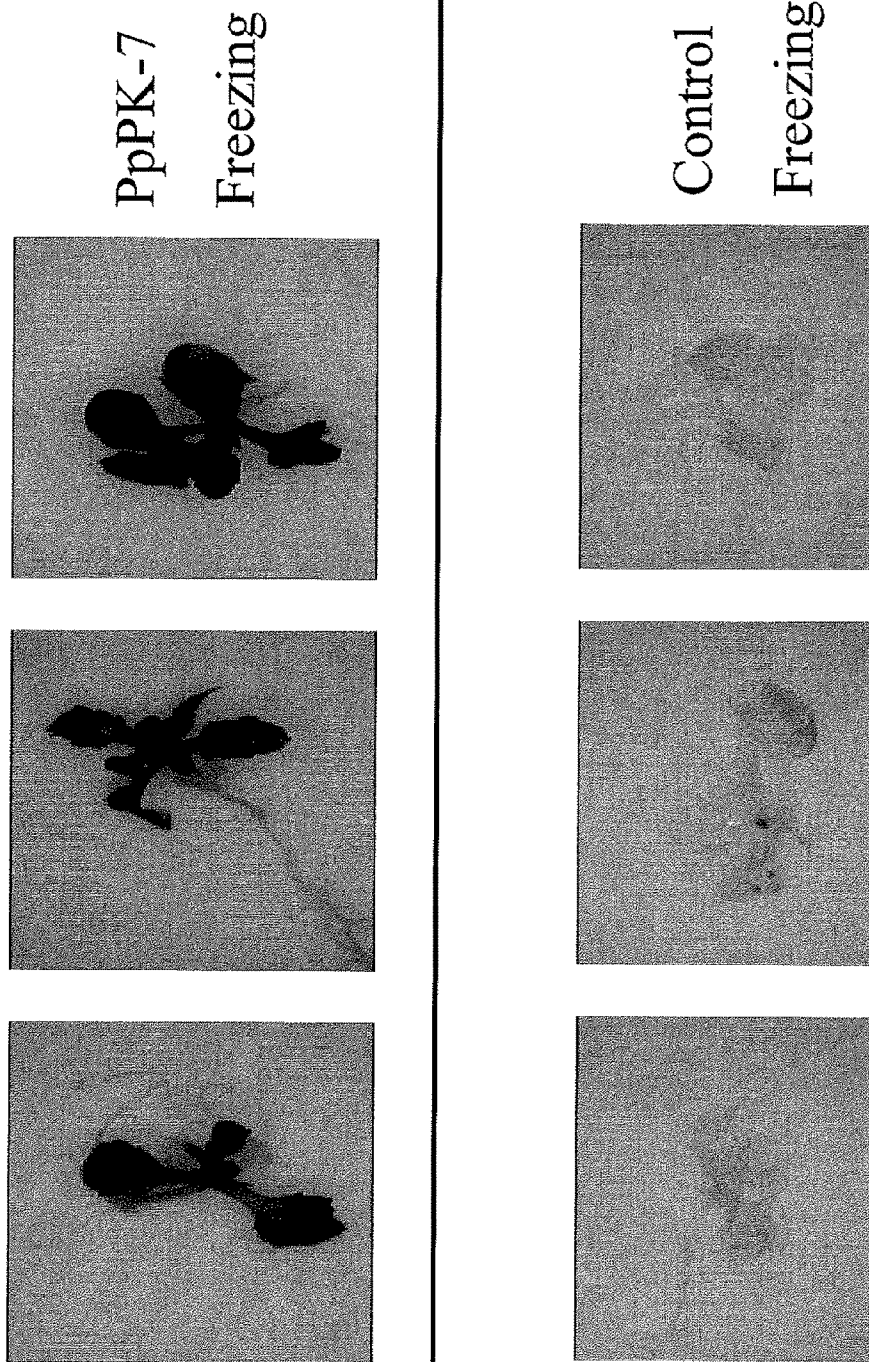


Figure 5

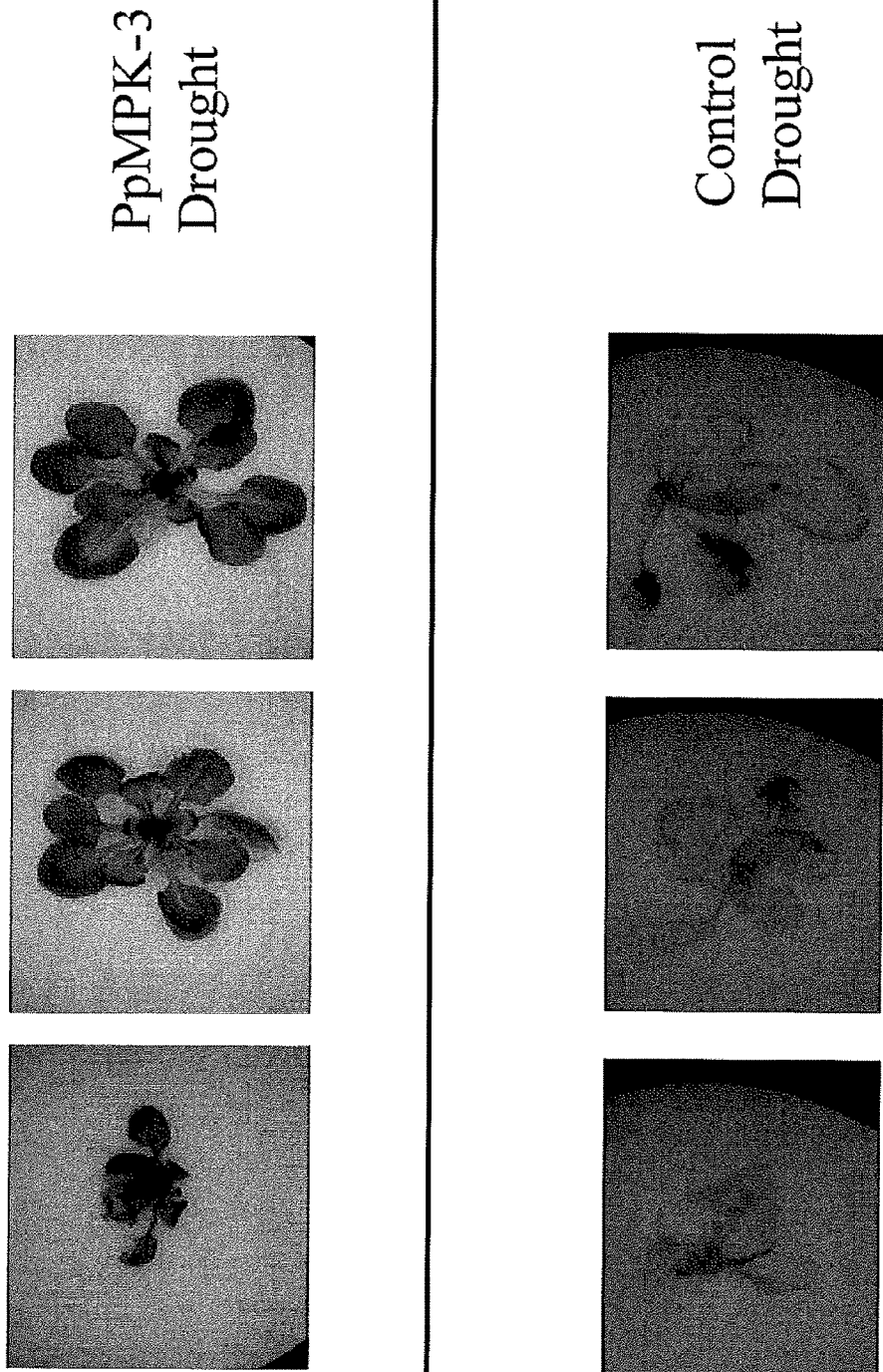


Figure 6

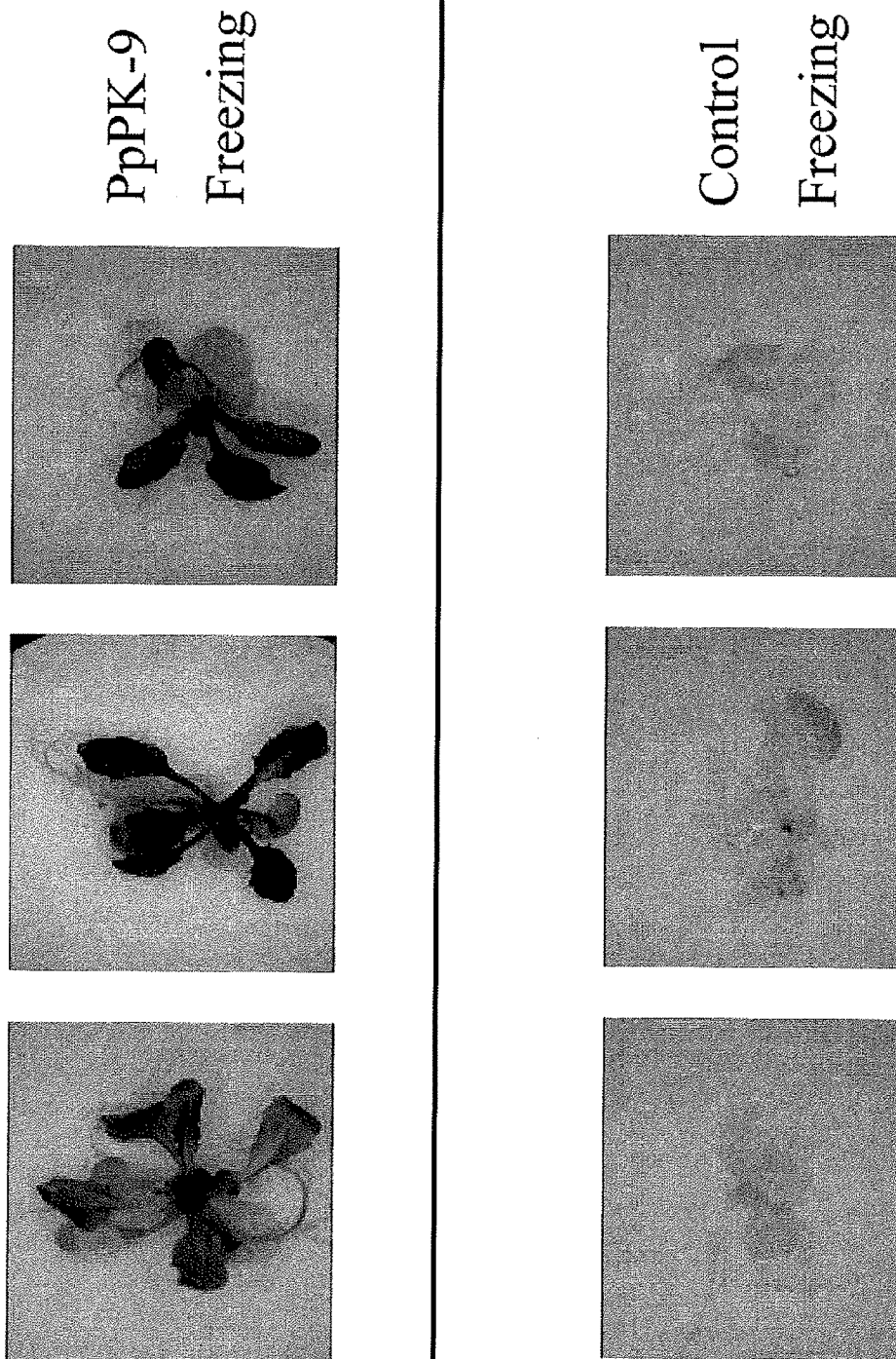


Figure 7

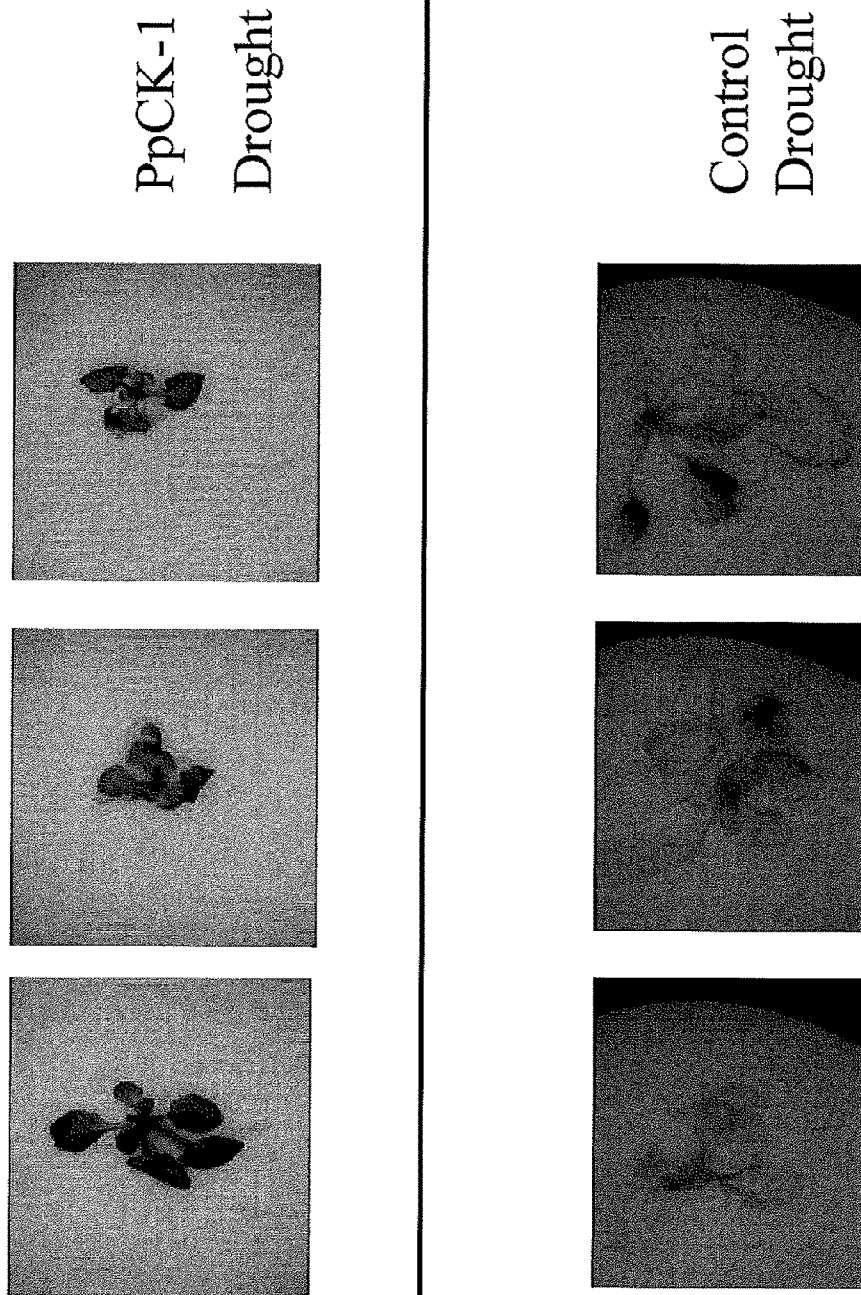




Figure 8

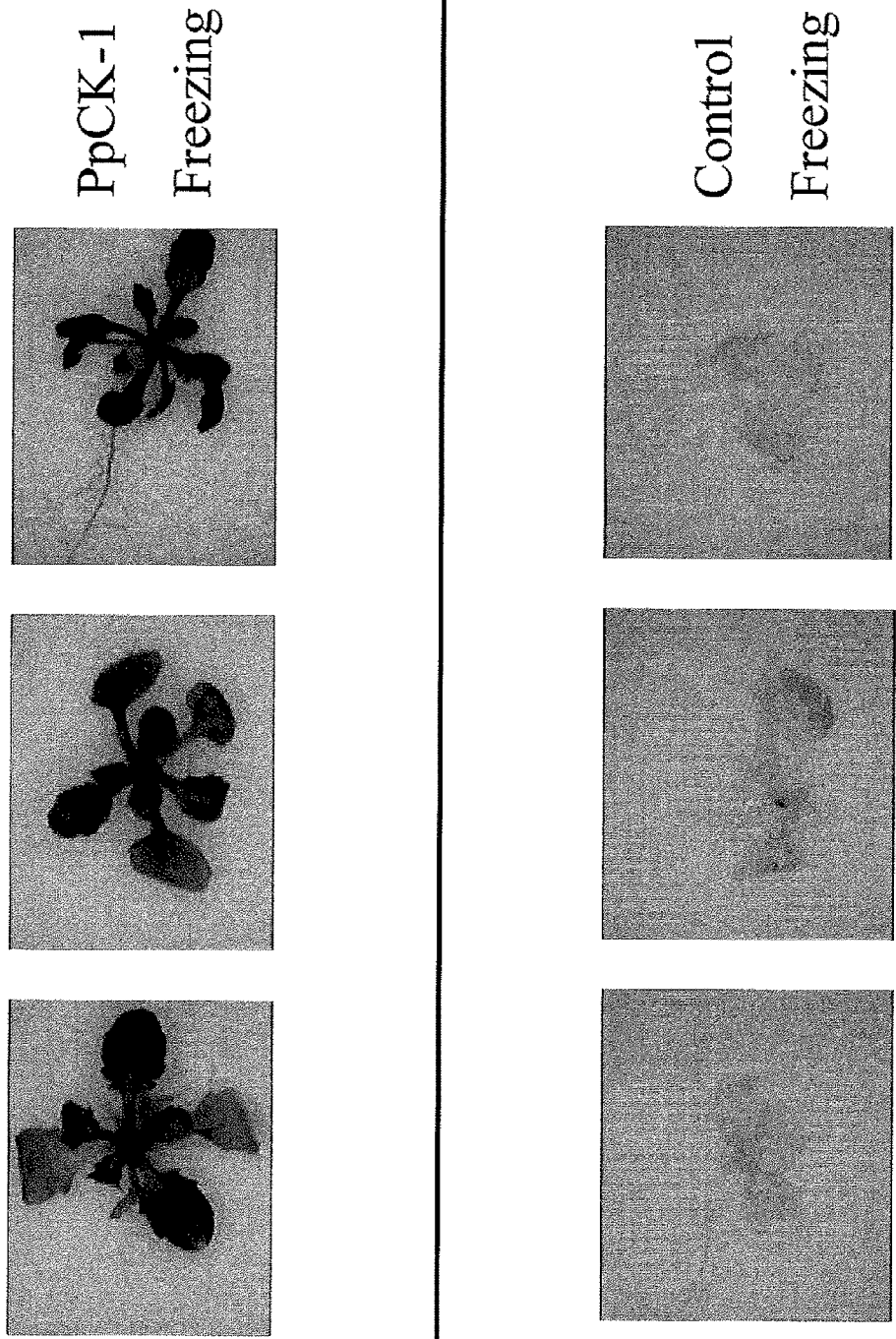


Figure 9

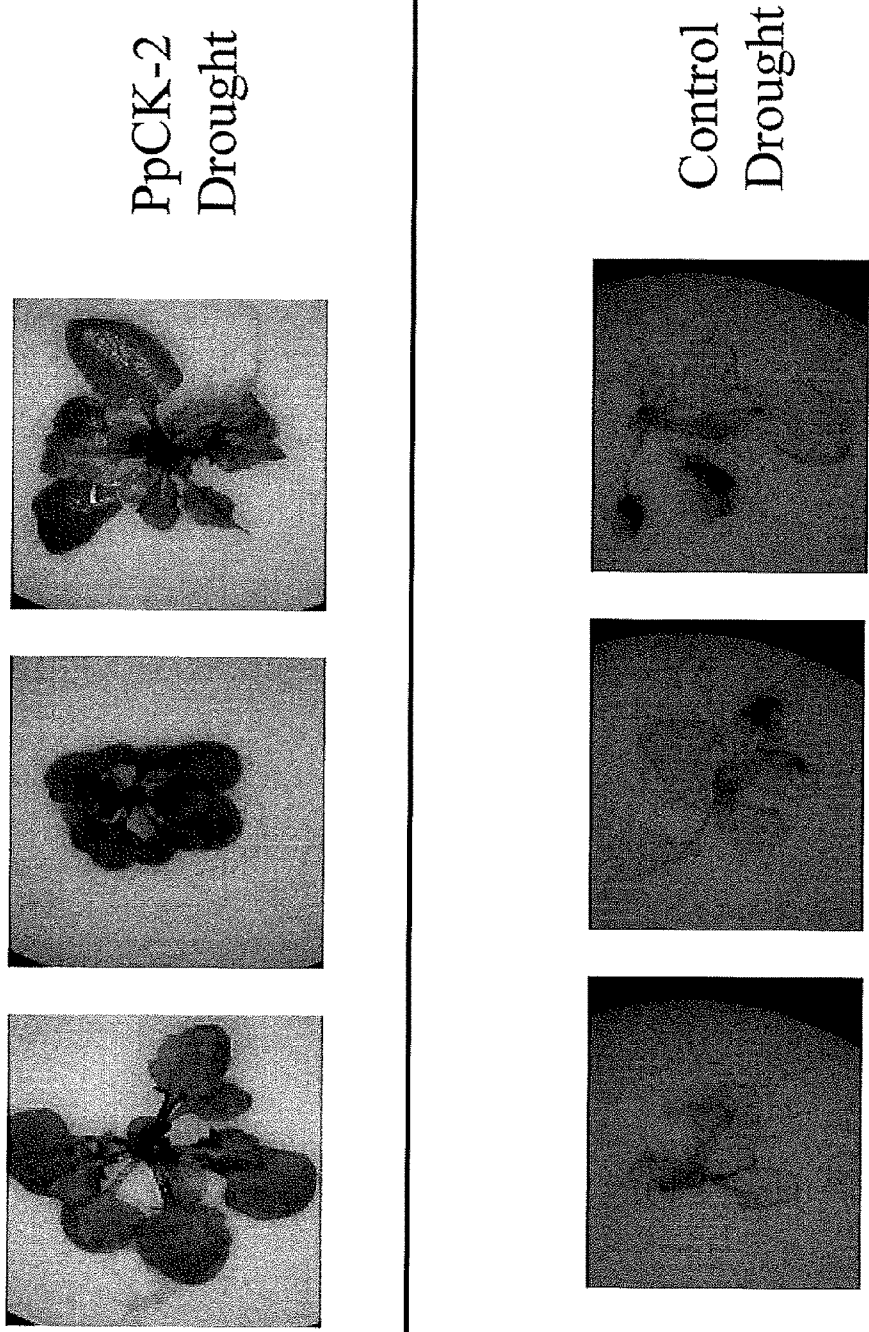


Figure 10

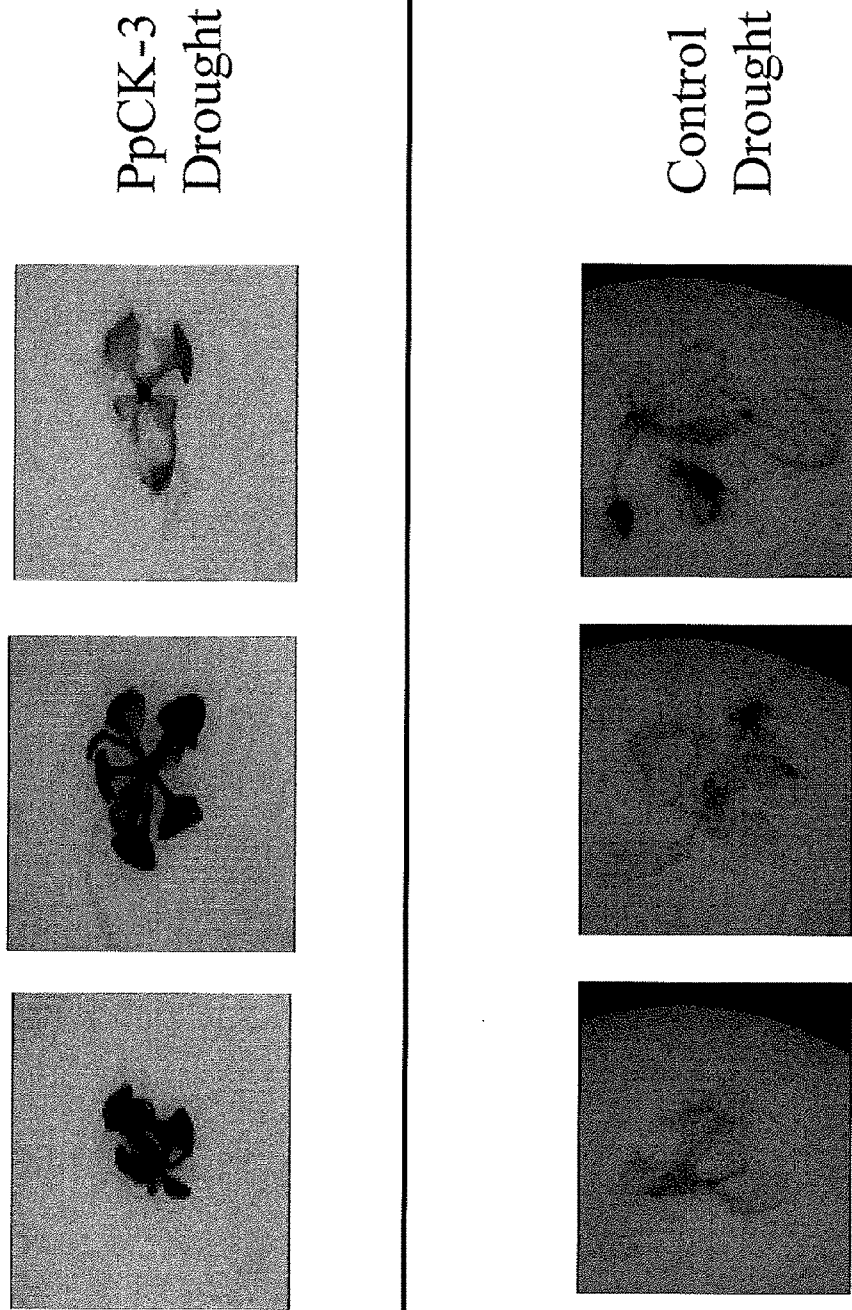


Figure 11

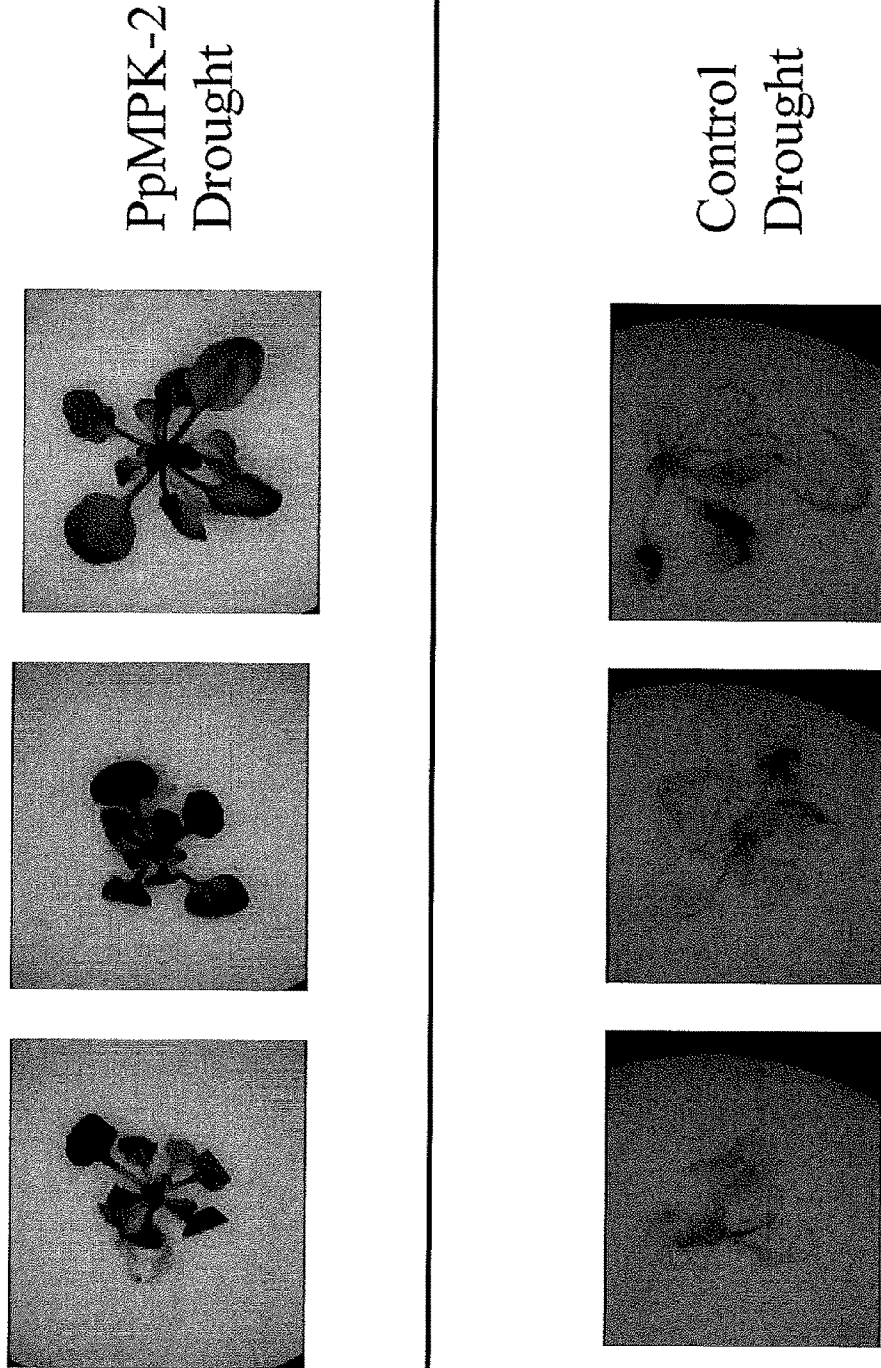


Figure 12

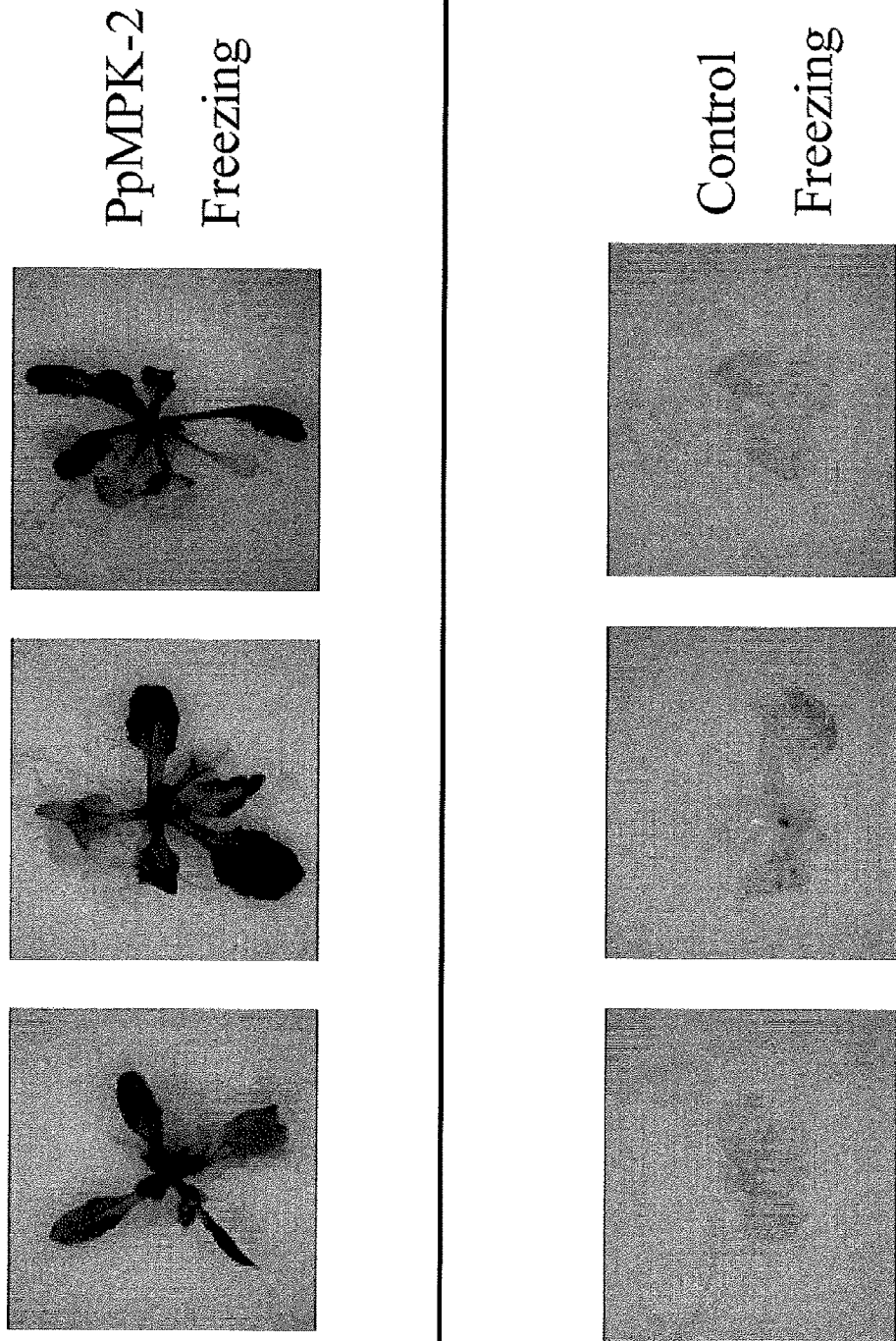


Figure 13

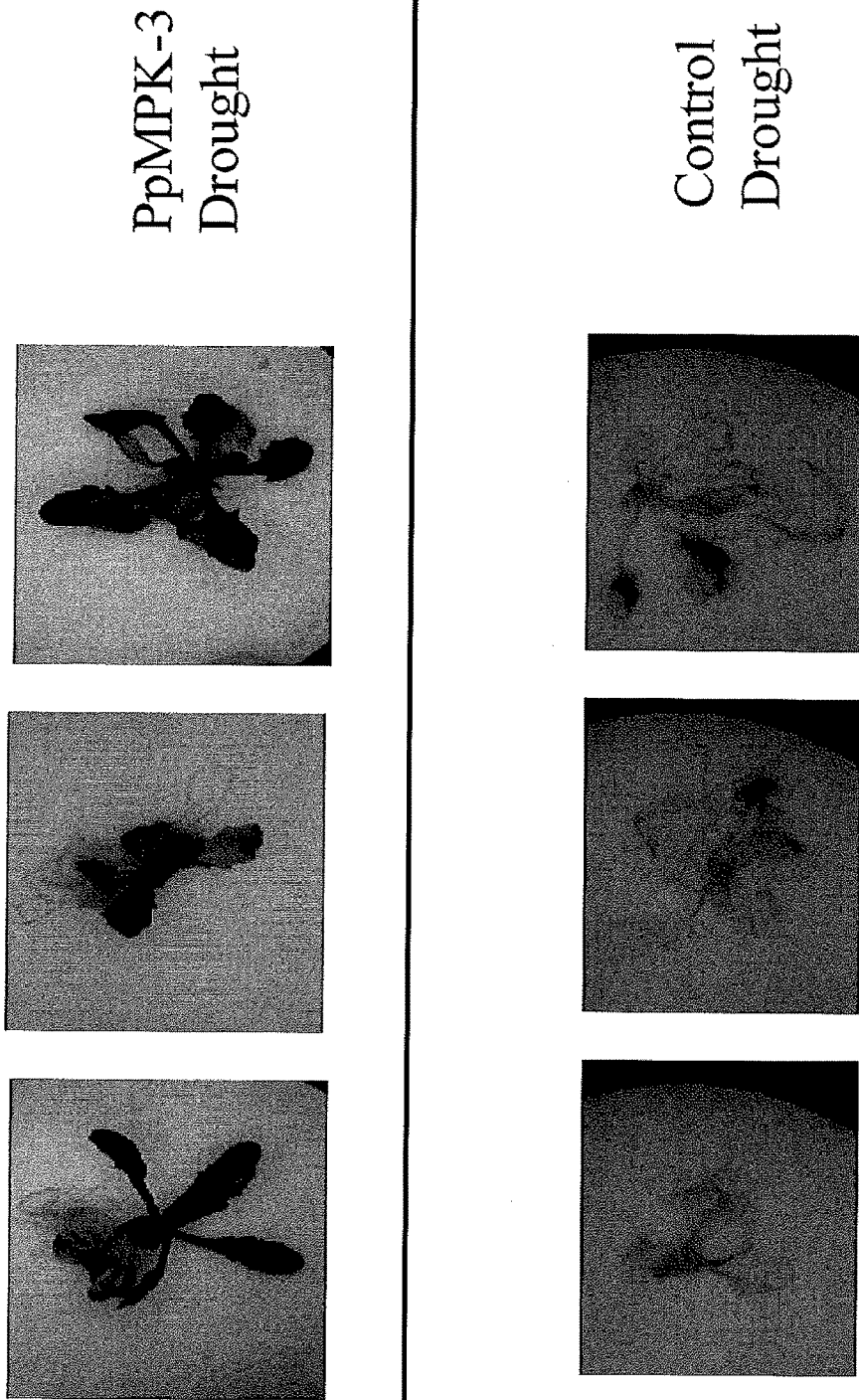


Figure 14

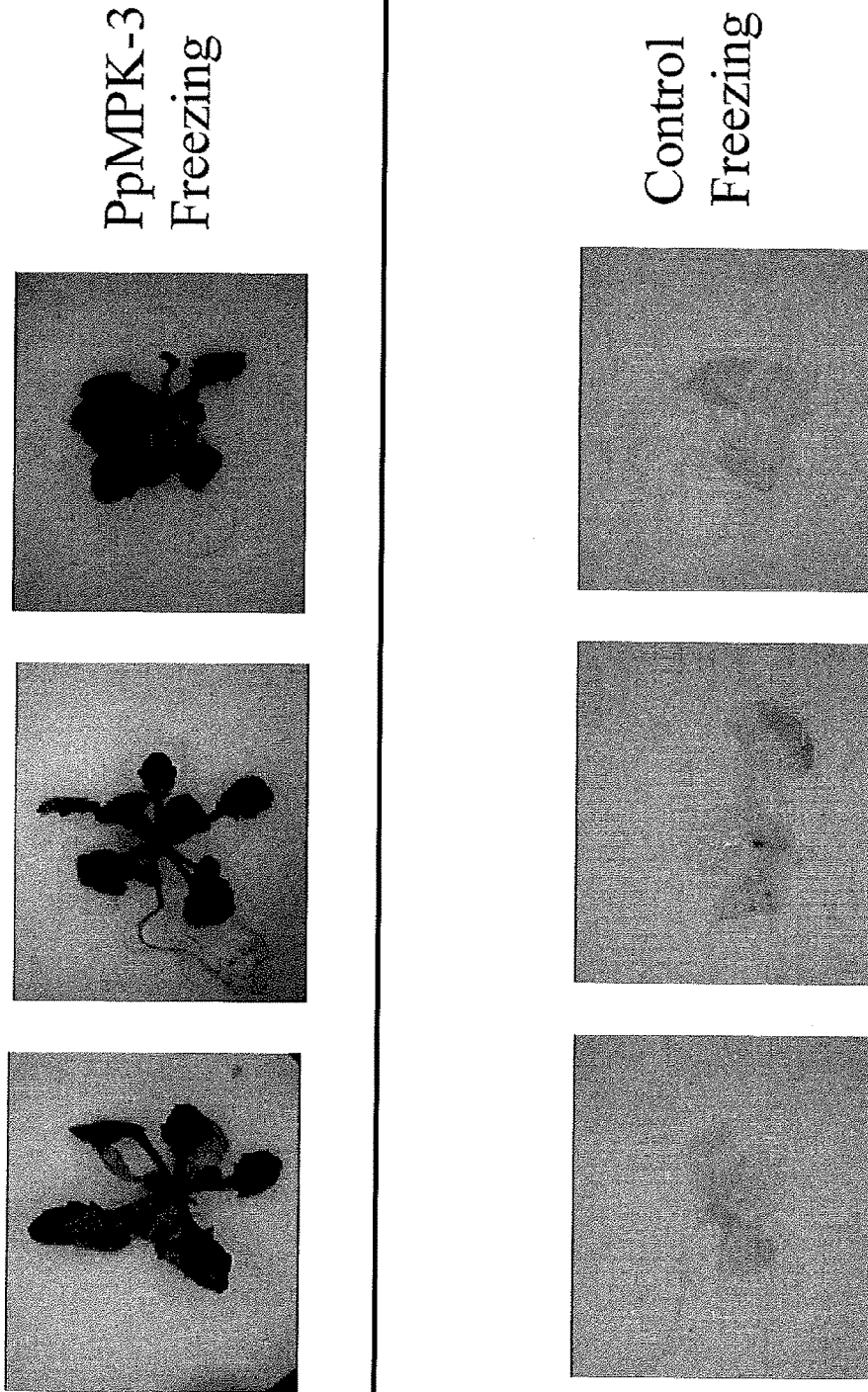


Figure 15

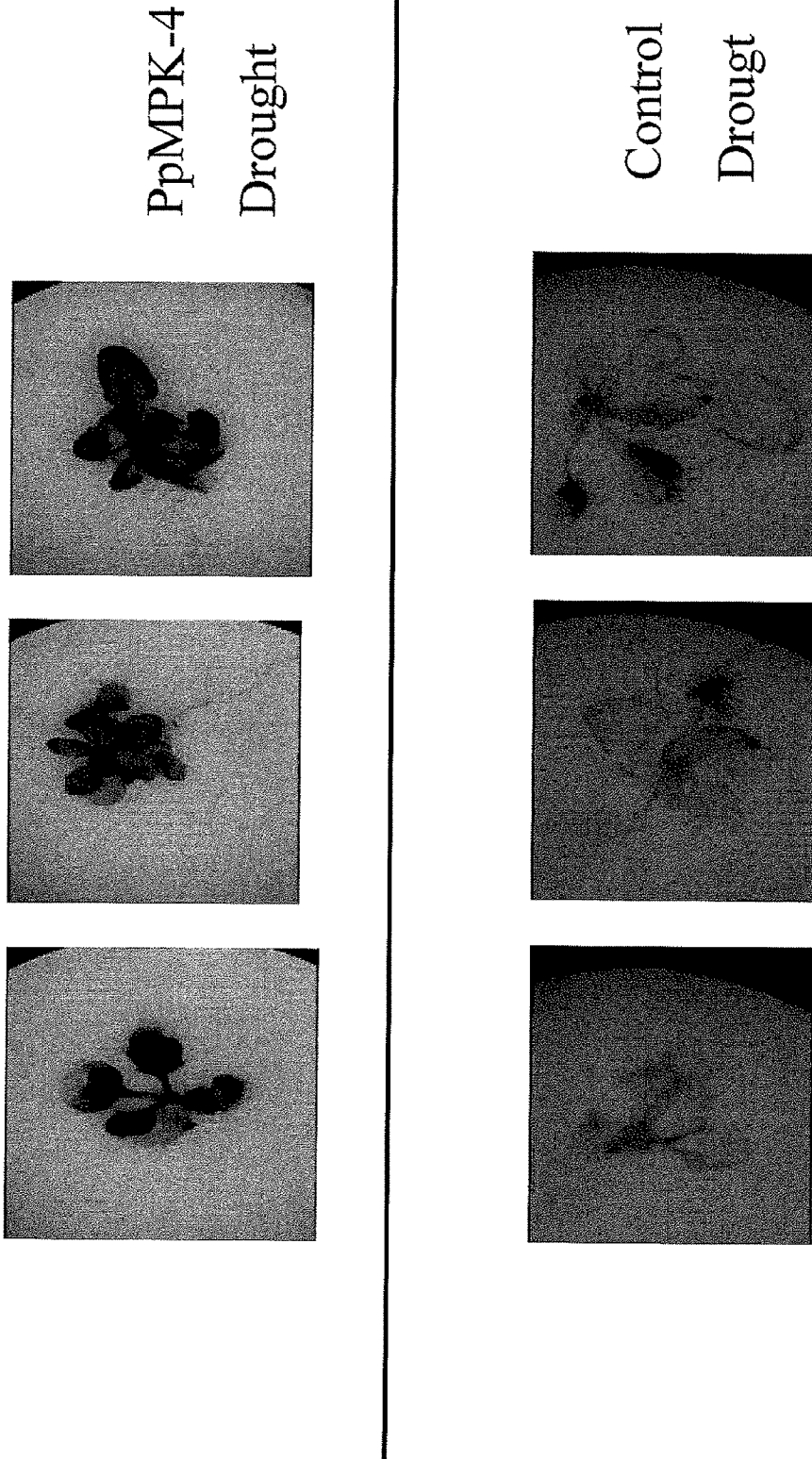




Figure 16

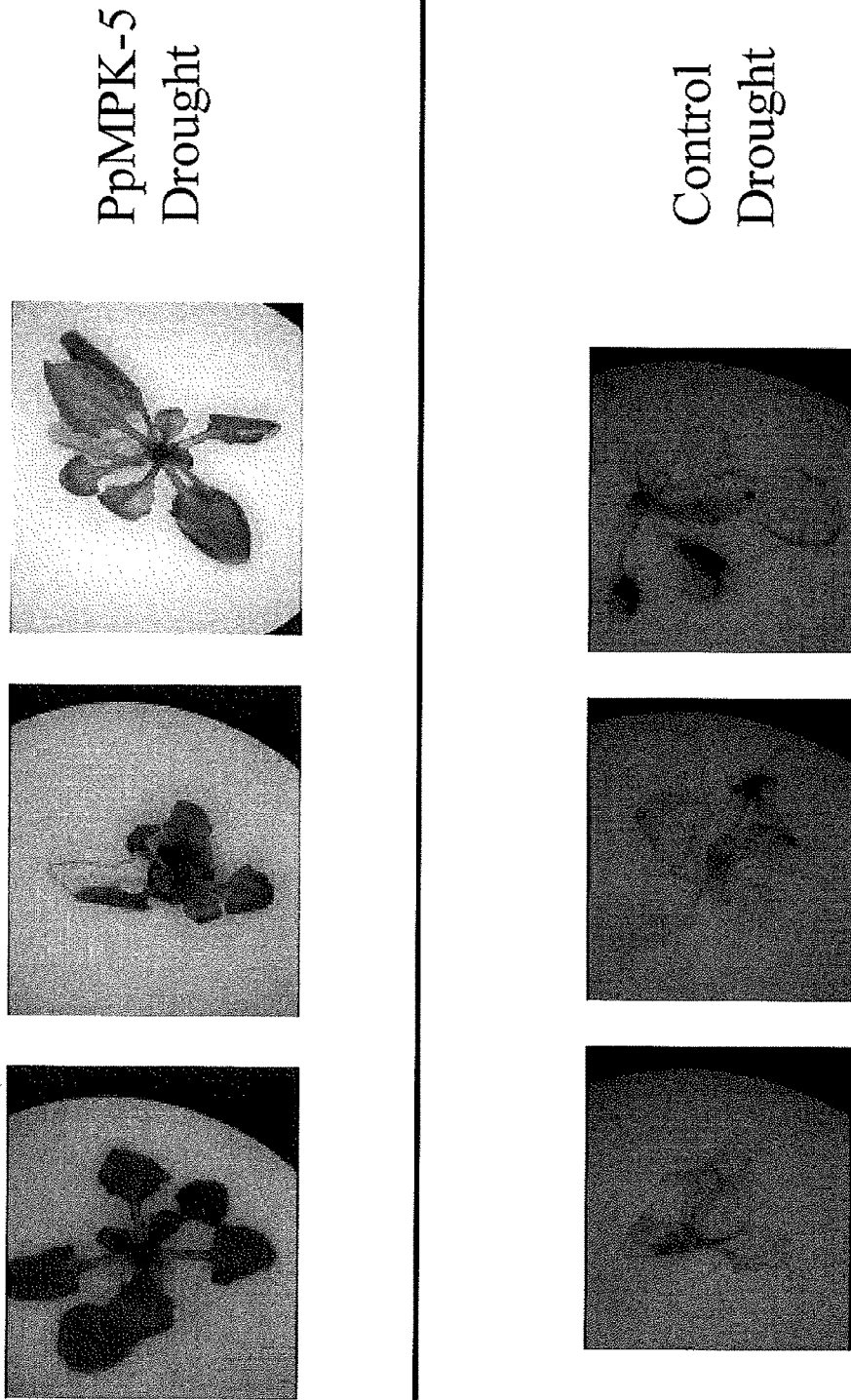


Figure 17

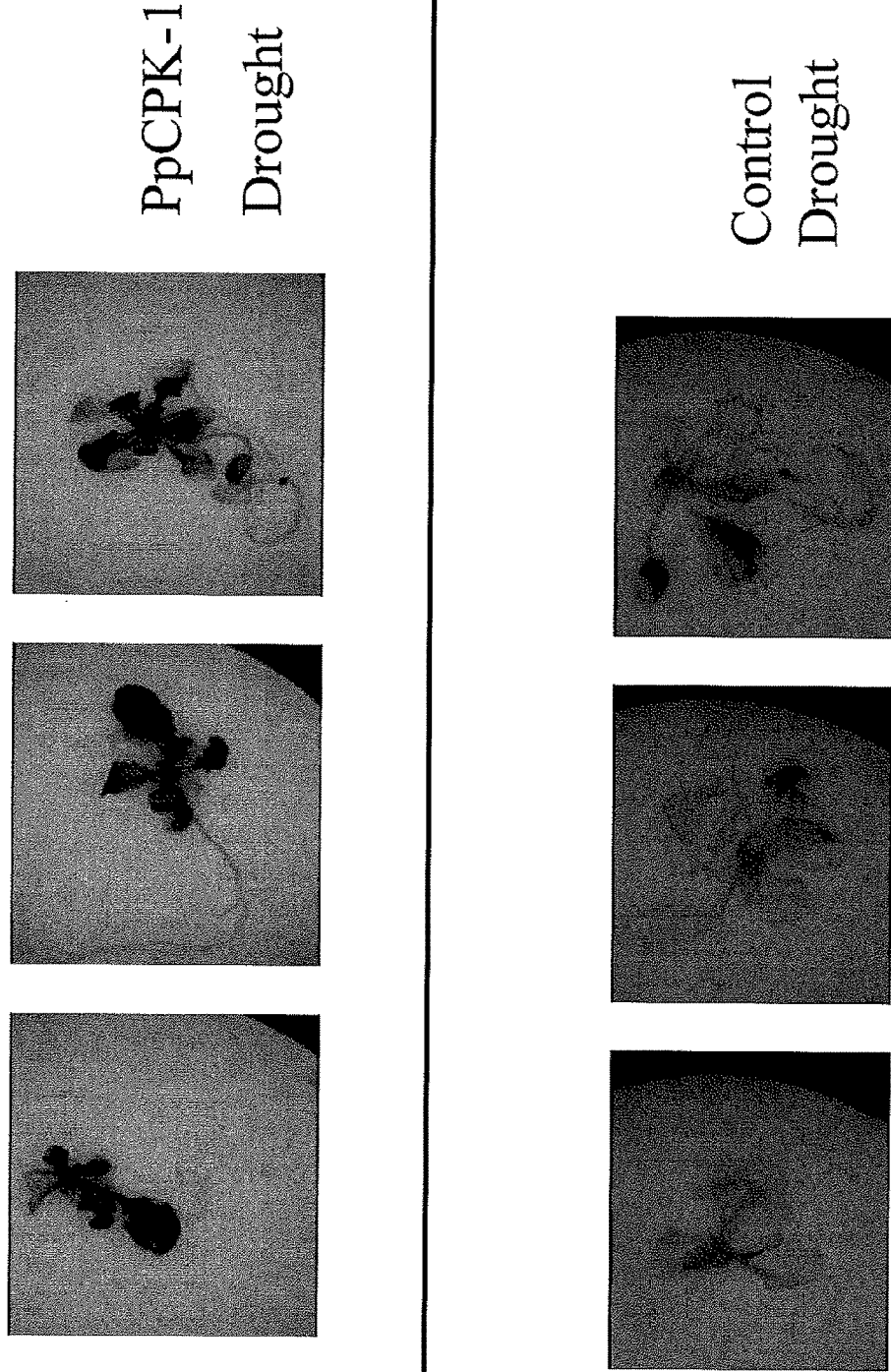
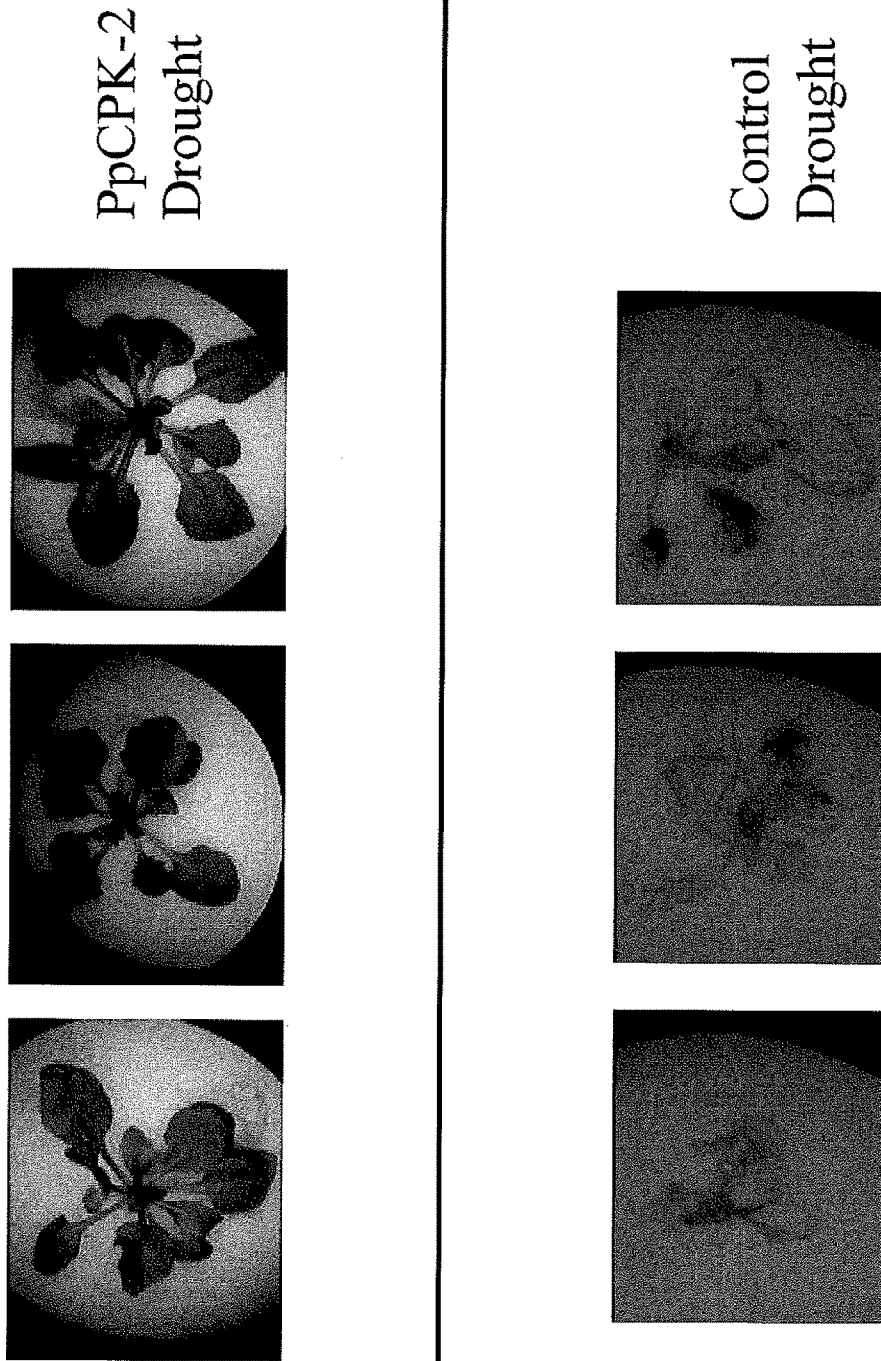


Figure 18



**PROTEIN KINASE STRESS-RELATED  
PROTEINS AND METHODS OF USE IN  
PLANTS**

CROSS REFERENCE TO RELATED  
APPLICATIONS

This application is a divisional of U.S. patent application Ser. No. 09/828,313, filed Apr. 6, 2001 and now U.S. Pat. No. 6,867,351, and is with U.S. patent application Ser. No. 12/545,903, filed Aug. 24, 2009, and is with U.S. patent application Ser. No. 12/401,635, filed Mar. 11, 2009, which is with U.S. patent application Ser. No. 11/807,408, filed May 29, 2007, and now U.S. Pat. No. 7,521,597, which is with U.S. patent application Ser. No. 11/961,634, filed Dec. 20, 2007, and now U.S. Pat. No. 7,521,598, which is a continuation of U.S. patent application Ser. No. 11/564,902, filed Nov. 30, 2006, and now U.S. Pat. No. 7,504,559, which is a continuation of U.S. patent application Ser. No. 10/768,863, filed Jan. 30, 2004 and now U.S. Pat. No. 7,179,962, which is a divisional of U.S. patent application Ser. No. 09/828,313, which claims the priority benefit of U.S. Provisional Application Ser. No. 60/196,001 filed Apr. 7, 2000. The contents of each of the above-identified applications are hereby incorporated by reference.

BACKGROUND OF THE INVENTION

1. Field of the Invention

This invention relates generally to nucleic acid sequences encoding proteins that are associated with abiotic stress responses and abiotic stress tolerance in plants. In particular, this invention relates to nucleic acid sequences encoding proteins that confer drought, cold, and/or salt tolerance to plants.

2. Background Art

Abiotic environmental stresses, such as drought stress, salinity stress, heat stress, and cold stress, are major limiting factors of plant growth and productivity. Crop losses and crop yield losses of major crops such as rice, maize (corn) and wheat caused by these stresses represent a significant economic and political factor and contribute to food shortages in many underdeveloped countries.

Plants are typically exposed during their life cycle to conditions of reduced environmental water content. Most plants have evolved strategies to protect themselves against these conditions of desiccation. However, if the severity and duration of the drought conditions are too great, the effects on plant development, growth and yield of most crop plants are profound. Furthermore, most of the crop plants are very susceptible to higher salt concentrations in the soil. Continuous exposure to drought and high salt causes major alterations in the plant metabolism. These great changes in metabolism ultimately lead to cell death and consequently yield losses.

Developing stress-tolerant plants is a strategy that has the potential to solve or mediate at least some of these problems. However, traditional plant breeding strategies to develop new lines of plants that exhibit resistance (tolerance) to these types of stresses are relatively slow and require specific resistant lines for crossing with the desired line. Limited germplasm resources for stress tolerance and incompatibility in crosses between distantly related plant species represent significant problems encountered in conventional breeding. Additionally, the cellular processes leading to drought, cold and salt tolerance in model, drought- and/or salt-tolerant plants are complex in nature and involve multiple mechanisms of cellular adaptation and numerous metabolic pathways. This multi-component nature of stress tolerance has not only made

breeding for tolerance largely unsuccessful, but has also limited the ability to genetically engineer stress tolerance plants using biotechnological methods.

Drought, cold as well as salt stresses have a common theme important for plant growth and that is water availability. Plants are exposed during their entire life cycle to conditions of reduced environmental water content. Most plants have evolved strategies to protect themselves against these conditions of desiccation. However, if the severity and duration of the drought conditions are too great, the effects on plant development, growth and yield of most crop plants are profound. Since high salt content in some soils result in less available water for cell intake, its effect is similar to those observed under drought conditions. Additionally, under freezing temperatures, plant cells lose water as a result of ice formation that starts in the apoplast and withdraws water from the symplast. Commonly, a plant's molecular response mechanisms to each of these stress conditions are common and protein kinases play an essential role in these molecular mechanisms.

Protein kinases represent a super family and the members of this family catalyze the reversible transfer of a phosphate group of ATP to serine, threonine and tyrosine amino acid side chains on target proteins. Protein kinases are primary elements in signaling processes in plants and have been reported to play crucial roles in perception and transduction of signals that allow a cell (and the plant) to respond to environmental stimuli. In particular, receptor protein kinases (RPKs) represent one group of protein kinases that activate a complex array of intracellular signaling pathways in response to the extracellular environment (Van der Gear et al., 1994 *Annu. Rev. Cell Biol.* 10:251-337). RPKs are single-pass transmembrane proteins that contain an amino-terminal signal sequence, extracellular domains unique to each receptor, and a cytoplasmic kinase domain. Ligand binding induces homo- or hetero-dimerization of RPKs, and the resultant close proximity of the cytoplasmic domains results in kinase activation by transphosphorylation. Although plants have many proteins similar to RPKs, no ligand has been identified for these receptor-like kinases (RLKs). The majority of plant RLKs that have been identified belong to the family of Serine/Threonine (Ser/Thr) kinases, and most have extracellular Leucine-rich repeats (Becraft, P W. 1998 *Trends Plant Sci.* 3:384-388).

Another type of protein kinase is the Ca<sup>+</sup>-dependent protein kinase (CDPK). This type of kinase has a calmodulin-like domain at the COOH terminus which allows response to Ca<sup>+</sup> signals directly without calmodulin being present. Currently, CDPKs are the most prevalent Ser/Thr protein kinases found in higher plants. Although their physiological roles remain unclear, they are induced by cold, drought and abscisic acid (ABA) (Knight et al., 1991 *Nature* 352:524; Schroeder, J I and Thuleau, P., 1991 *Plant Cell* 3:555; Bush, D. S., 1995 *Annu. Rev. Plant Phys. Plant Mol. Biol.* 46:95; Urao, T. et al., 1994 *Mol. Gen. Genet.* 244:331).

Another type of signaling mechanism involves members of the conserved SNF1 Serine/Threonine protein kinase family. These kinases play essential roles in eukaryotic glucose and stress signaling. Plant SNF1-like kinases participate in the control of key metabolic enzymes, including HMGR, nitrate reductase, sucrose synthase, and sucrose phosphate synthase (SPS). Genetic and biochemical data indicate that sugar-dependent regulation of SNF1 kinases involves several other sensory and signaling components in yeast, plants and animals.

Additionally, members of the Mitogen-Activated Protein Kinase (MAPK) family have been implicated in the actions of

numerous environmental stresses in animals, yeasts and plants. It has been demonstrated that both MAPK-like kinase activity and mRNA levels of the components of MAPK cascades increase in response to environmental stress and plant hormone signal transduction. MAP kinases are components of sequential kinase cascades, which are activated by phosphorylation of threonine and tyrosine residues by intermediate upstream MAP kinase kinases (MAPKKs). The MAPKKs are themselves activated by phosphorylation of serine and threonine residues by upstream kinases (MAPKKKs). A number of MAP Kinase genes have been reported in higher plants.

#### SUMMARY OF THE INVENTION

This invention fulfills in part the need to identify new, unique protein kinases capable of conferring stress tolerance to plants upon over-expression. The present invention provides a transgenic plant cell transformed by a Protein Kinase Stress-Related Protein (PKSRP) coding nucleic acid, wherein expression of the nucleic acid sequence in the plant cell results in increased tolerance to environmental stress as compared to a wild type variety of the plant cell. Namely, described herein are the protein kinases: 1) Ser/Thr Kinase and other type of kinases (PK-6, PK-7, PK-8 and PK-9); 2) Calcium dependent protein kinases (CDPK-1 and CDPK-2), 3) Casein Kinase homologs (CK-1, CK-2 and CK-3), and 4) MAP-Kinases (MPK-2, MPK-3, MPK-4 and MPK-5), all from *Physcomitrella patens*.

The invention provides in some embodiments that the PKSRP and coding nucleic acid are that found in members of the genus *Physcomitrella*. In another preferred embodiment, the nucleic acid and protein are from a *Physcomitrella patens*. The invention provides that the environmental stress can be salinity, drought, temperature, metal, chemical, pathogenic and oxidative stresses, or combinations thereof. In preferred embodiments, the environmental stress can be drought or cold temperature.

The invention further provides a seed produced by a transgenic plant transformed by a PKSRP coding nucleic acid, wherein the plant is true breeding for increased tolerance to environmental stress as compared to a wild type variety of the plant. The invention further provides a seed produced by a transgenic plant expressing a PKSRP, wherein the plant is true breeding for increased tolerance to environmental stress as compared to a wild type variety of the plant.

The invention further provides an agricultural product produced by any of the below-described transgenic plants, plant parts or seeds. The invention further provides an isolated PKSRP as described below. The invention further provides an isolated PKSRP coding nucleic acid, wherein the PKSRP coding nucleic acid codes for a PKSRP as described below.

The invention further provides an isolated recombinant expression vector comprising a PKSRP coding nucleic acid as described below, wherein expression of the vector in a host cell results in increased tolerance to environmental stress as compared to a wild type variety of the host cell. The invention further provides a host cell containing the vector and a plant containing the host cell.

The invention further provides a method of producing a transgenic plant with a PKSRP coding nucleic acid, wherein expression of the nucleic acid in the plant results in increased tolerance to environmental stress as compared to a wild type variety of the plant comprising: (a) transforming a plant cell with an expression vector comprising a PKSRP coding nucleic acid, and (b) generating from the plant cell a transgenic plant with an increased tolerance to environmental

stress as compared to a wild type variety of the plant. In preferred embodiments, the PKSRP and PKSRP coding nucleic acid are as described below.

The present invention further provides a method of identifying a novel PKSRP, comprising (a) raising a specific antibody response to a PKSRP, or fragment thereof, as described below; (b) screening putative PKSRP material with the antibody, wherein specific binding of the antibody to the material indicates the presence of a potentially novel PKSRP; and (c) identifying from the bound material a novel PKSRP in comparison to known PKSRP. Alternatively, hybridization with nucleic acid probes as described below can be used to identify novel PKSRP nucleic acids.

The present invention also provides methods of modifying stress tolerance of a plant comprising, modifying the expression of a PKSRP nucleic acid in the plant, wherein the PKSRP is as described below. The invention provides that this method can be performed such that the stress tolerance is either increased or decreased. Preferably, stress tolerance is increased in a plant via increasing expression of a PKSRP nucleic acid.

#### BRIEF DESCRIPTION OF THE DRAWINGS

FIG. 1 shows a diagram of the plant expression vector pBPSSC022 containing the super promoter driving the expression of SEQ ID NOs: 14, 15, 16, 17, 18, 19, 20, 21, 22, 23, 24, 25 and 26 ("Desired Gene"). The components are: NPTII kanamycin resistance gene (Bevan M, Nucleic Acids Res. 26: 8711-21, 1984), AtAct2-i promoter (An Y Q et al., Plant J 10: 107-121 1996), OCS3 terminator (During K, Transgenic Res. 3: 138-140, 1994), NOSpA terminator (Jefferson et al., EMBO J 6:3901-7 1987).

FIG. 2 shows the results of a drought stress test with over-expressing PpPK-6 transgenic plants and wild-type *Arabidopsis* lines. The transgenic lines display a tolerant phenotype. Individual transformant lines are shown.

FIG. 3 shows the results of a drought stress test with over-expressing PpPK-7 transgenic plants and wild-type *Arabidopsis* lines. The transgenic lines display a tolerant phenotype. Individual transformant lines are shown.

FIG. 4 shows the results of a freezing stress test with over-expressing PpPK-7 transgenic plants and wild-type *Arabidopsis* lines. The transgenic lines display a tolerant phenotype. Individual transformant lines are shown.

FIG. 5 shows the results of a drought stress test with over-expressing PpPK-9 transgenic plants and wild-type *Arabidopsis* lines. The transgenic lines display a tolerant phenotype. Individual transformant lines are shown.

FIG. 6 shows the results of a freezing stress test with over-expressing PpPK-9 transgenic plants and wild-type *Arabidopsis* lines. The transgenic lines display a tolerant phenotype. Individual transformant lines are shown.

FIG. 7 shows the results of a drought stress test with over-expressing PpCK-1 transgenic plants and wild-type *Arabidopsis* lines. The transgenic lines display a tolerant phenotype. Individual transformant lines are shown.

FIG. 8 shows the results of a freezing stress test with over-expressing PpCK-1 transgenic plants and wild-type *Arabidopsis* lines. The transgenic lines display a tolerant phenotype. Individual transformant lines are shown.

FIG. 9 shows the results of a drought stress test with over-expressing PpCK-2 transgenic plants and wild-type *Arabidopsis* lines. The transgenic lines display a tolerant phenotype. Individual transformant lines are shown.

FIG. 10 shows the results of a drought stress test with over-expressing PpCK-3 transgenic plants and wild-type

*Arabidopsis* lines. The transgenic lines display a tolerant phenotype. Individual transformant lines are shown.

FIG. 11 shows the results of a drought stress test with over-expressing PpMPK-2 transgenic plants and wild-type *Arabidopsis* lines. The transgenic lines display a tolerant phenotype. Individual transformant lines are shown.

FIG. 12 shows the results of a freezing stress test with over-expressing PpMPK-2 transgenic plants and wild-type *Arabidopsis* lines. The transgenic lines display a tolerant phenotype. Individual transformant lines are shown.

FIG. 13 shows the results of a drought stress test with over-expressing PpMPK-3 transgenic plants and wild-type *Arabidopsis* lines. The transgenic lines display a tolerant phenotype. Individual transformant lines are shown.

FIG. 14 shows the results of a freezing stress test with over-expressing PpMPK-3 transgenic plants and wild-type *Arabidopsis* lines. The transgenic lines display a tolerant phenotype. Individual transformant lines are shown.

FIG. 15 shows the results of a drought stress test with over-expressing PpMPK-4 transgenic plants and wild-type *Arabidopsis* lines. The transgenic lines display a tolerant phenotype. Individual transformant lines are shown.

FIG. 16 shows the results of a drought stress test with over-expressing PpMPK-5 transgenic plants and wild-type *Arabidopsis* lines. The transgenic lines display a tolerant phenotype. Individual transformant lines are shown.

FIG. 17 shows the results of a drought stress test with over-expressing PpCPK-1 transgenic plants and wild-type *Arabidopsis* lines. The transgenic lines display a tolerant phenotype. Individual transformant lines are shown.

FIG. 18 shows the results of a drought stress test with over-expressing PpCPK-2 transgenic plants and wild-type *Arabidopsis* lines. The transgenic lines display a tolerant phenotype. Individual transformant lines are shown.

#### DETAILED DESCRIPTION OF THE INVENTION

The present invention may be understood more readily by reference to the following detailed description of the preferred embodiments of the invention and the Examples included herein. However, before the present compounds, compositions, and methods are disclosed and described, it is to be understood that this invention is not limited to specific nucleic acids, specific polypeptides, specific cell types, specific host cells, specific conditions, or specific methods, etc., as such may, of course, vary, and the numerous modifications and variations therein will be apparent to those skilled in the art. It is also to be understood that the terminology used herein is for the purpose of describing specific embodiments only and is not intended to be limiting. In particular, the designation of the amino acid sequences as protein "Protein Kinase Stress-Related Proteins" (PKSRPs), in no way limits the functionality of those sequences.

The present invention provides a transgenic plant cell transformed by a PKSRP coding nucleic acid, wherein expression of the nucleic acid sequence in the plant cell results in increased tolerance to environmental stress as compared to a wild type variety of the plant cell. The invention further provides transgenic plant pails and transgenic plants containing the plant cells described herein. Also provided is a plant seed produced by a transgenic plant transformed by a PKSRP coding nucleic acid, wherein the seed contains the PKSRP coding nucleic acid, and wherein the plant is true breeding for increased tolerance to environmental stress as compared to a wild type variety of the plant. The invention further provides a seed produced by a transgenic plant expressing a PKSRP, wherein the seed contains the PKSRP,

and wherein the plant is true breeding for increased tolerance to environmental stress as compared to a wild type variety of the plant. The invention also provides an agricultural product produced by any of the below-described transgenic plants, plant parts and plant seeds.

As used herein, the term "variety" refers to a group of plants within a species that share constant characters that separate them from the typical form and from other possible varieties within that species. While possessing at least one distinctive trait, a variety is also characterized by some variation between individuals within the variety, based primarily on the Mendelian segregation of traits among the progeny of succeeding generations. A variety is considered "true breeding" for a particular trait if it is genetically homozygous for that trait to the extent that when the true-breeding variety is self-pollinated, a significant amount of independent segregation of the trait among the progeny is not observed. In the present invention, the trait arises from the transgenic expression of one or more DNA sequences introduced into a plant variety.

The present invention describes for the first time that the *Physcomitrella patens* PKSRPs, PK-6, PK-7, PK-8, PK-9, CK-1, CK-2, CK-3, MPK-2, MPK-3, MPK-4, MPK-5, CPK-1 and CPK-2, are useful for increasing a plant's tolerance to environmental stress. Accordingly, the present invention provides isolated PKSRPs selected from the group consisting of PK-6, PK-7, PK-8, PK-9, CK-1, CK-2, CK-3, MPK-2, MPK-3, MPK-4, MPK-5, CPK-1 and CPK-2, and homologs thereof. In preferred embodiments, the PKSRP is selected from 1) Protein Kinase-6 (PK-6) protein as defined in SEQ ID NO:27; 2) Protein Kinase-7 (PK-7) protein as defined in SEQ ID NO:28; 3) Protein Kinase-8 (PK-8) protein as defined in SEQ ID NO:29; 4) Protein Kinase-9 (PK-9) protein as defined in SEQ ID NO:30; 5) Casein Kinase homologue (CK-1) protein as defined in SEQ ID NO:31; 6) Casein Kinase homologue-2 (CK-2) protein as defined in SEQ ID NO:32; 7) Casein Kinase homologue-3 (CK-3) protein as defined in SEQ ID NO:33; 8) MAP Kinase-2 (MPK-2) protein as defined in SEQ ID NO:34; 9) MAP Kinase-3 (MPK-3) protein as defined in SEQ ID NO:35; 10) MAP Kinase-4 (MPK-4) protein as defined in SEQ ID NO:36; 11) MAP Kinase-5 (MPK-5) protein as defined in SEQ ID NO:37; 12) Calcium dependent protein kinase-1 (CPK-1) protein as defined in SEQ ID NO:38; 13) Calcium dependent protein kinase-2 (CPK-2) protein as defined in SEQ ID NO:39; and homologs and orthologs thereof. Homologs and orthologs of the amino acid sequences are defined below.

The PKSRPs of the present invention are preferably produced by recombinant DNA techniques. For example, a nucleic acid molecule encoding the protein is cloned into an expression vector (as described below), the expression vector is introduced into a host cell (as described below) and the PKSRP is expressed in the host cell. The PKSRP can then be isolated from the cells by an appropriate purification scheme using standard protein purification techniques. Alternative to recombinant expression, a PKSRP polypeptide, or peptide can be synthesized chemically using standard peptide synthesis techniques. Moreover, native PKSRP can be isolated from cells (e.g., *Physcomitrella patens*), for example using an anti-PKSRP antibody, which can be produced by standard techniques utilizing a PKSRP or fragment thereof.

The invention further provides an isolated PKSRP coding nucleic acid. The present invention includes PKSRP coding nucleic acids that encode PKSRPs as described herein. In preferred embodiments, the PKSRP coding nucleic acid is selected from 1) Protein Kinase-6 (PK-6) nucleic acid as defined in SEQ ID NO:14; 2) Protein Kinase-7 (PK-7)

nucleic acid as defined in SEQ ID NO:15; 3) Protein Kinase-8 (PK-8) nucleic acid as defined in SEQ ID NO:16; 4) Protein Kinase-9 (PK-9) nucleic acid as defined in SEQ ID NO:17; 5) Casein Kinase homolog (CK-1) nucleic acid as defined in SEQ ID NO:18; 6) Casein Kinase homolog-2 (CK-2) nucleic acid as defined in SEQ ID NO:19; 7) Casein Kinase homolog-3 (CK-3) nucleic acid as defined in SEQ ID NO:20; 8) MAP Kinase-2 (MPK-2) nucleic acid as defined in SEQ ID NO:21; 9) MAP Kinase-3 (MPK-3) nucleic acid as defined in SEQ ID NO:22; 10) MAP Kinase-4 (MPK-4) nucleic acid as defined in SEQ ID NO:23; 11) MAP Kinase-5 (MPK-5) nucleic acid as defined in SEQ ID NO:24; 12) Calcium dependent protein kinase-1 (CPK-1) nucleic acid as defined in SEQ ID NO:25; 13) Calcium dependent protein kinase-2 (CPK-2) nucleic acid as defined in SEQ ID NO:26 and homologs and orthologs thereof Homologs and orthologs of the nucleotide sequences are defined below. In one preferred embodiment, the nucleic acid and protein are isolated from the plant genus *Physcomitrella*. In another preferred embodiment, the nucleic acid and protein are from a *Physcomitrella patens* (*P. patens*) plant.

As used herein, the term "environmental stress" refers to any sub-optimal growing condition and includes, but is not limited to, sub-optimal conditions associated with salinity, drought, temperature, metal, chemical, pathogenic and oxidative stresses, or combinations thereof. In preferred embodiments, the environmental stress can be salinity, drought, or temperature, or combinations thereof, and in particular, can be high salinity, low water content or low temperature. It is also to be understood that as used in the specification and in the claims, "a" or "an" can mean one or more, depending upon the context in which it is used. Thus, for example, reference to "a cell" can mean that at least one cell can be utilized.

As also used herein, the terms "nucleic acid" and "nucleic acid molecule" are intended to include DNA molecules (e.g., cDNA or genomic DNA) and RNA molecules (e.g., mRNA) and analogs of the DNA or RNA generated using nucleotide analogs. This term also encompasses untranslated sequence located at both the 3' and 5' ends of the coding region of the gene: at least about 1000 nucleotides of sequence upstream from the 5' end of the coding region and at least about 200 nucleotides of sequence downstream from the 3' end of the coding region of the gene. The nucleic acid molecule can be single-stranded or double-stranded, but preferably is double-stranded DNA.

An "isolated" nucleic acid molecule is one that is substantially separated from other nucleic acid molecules which are present in the natural source of the nucleic acid. Preferably, an "isolated" nucleic acid is free of some of the sequences which naturally flank the nucleic acid (i.e., sequences located at the 5' and 3' ends of the nucleic acid) in the genomic DNA of the organism from which the nucleic acid is derived. For example, in various embodiments, the isolated PKSRP nucleic acid molecule can contain less than about 5 kb, 4 kb, 3 kb, 2 kb, 1 kb, 0.5 kb or 0.1 kb of nucleotide sequences which naturally flank the nucleic acid molecule in genomic DNA of the cell from which the nucleic acid is derived (e.g., a *Physcomitrella patens* cell). Moreover, an "isolated" nucleic acid molecule, such as a cDNA molecule, can be free from some of the other cellular material with which it is naturally associated, or culture medium when produced by recombinant techniques, or chemical precursors or other chemicals when chemically synthesized.

A nucleic acid molecule of the present invention, e.g., a nucleic acid molecule having a nucleotide sequence of SEQ ID NO:14, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17,

SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24, SEQ ID NO:25 or SEQ ID NO:26, or a portion thereof, can be isolated using standard molecular biology techniques and the sequence information provided herein. For example, a *P. patens* PKSRP cDNA can be isolated from a *P. patens* library using all or portion of one of the sequences of SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:3, SEQ ID NO:4, SEQ ID NO:5, SEQ ID NO:6, SEQ ID NO:7, SEQ ID NO:8, SEQ ID NO:9, SEQ ID NO:10, SEQ ID NO:11, SEQ ID NO:12 and SEQ ID NO:13. Moreover, a nucleic acid molecule encompassing all or a portion of one of the sequences of SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:3, SEQ ID NO:4, SEQ ID NO:5, SEQ ID NO:6, SEQ ID NO:7, SEQ ID NO:8, SEQ ID NO:9, SEQ ID NO:10, SEQ ID NO:11, SEQ ID NO:12 and SEQ ID NO:13 can be isolated by the polymerase chain reaction using oligonucleotide primers designed based upon this sequence. For example, mRNA can be isolated from plant cells (e.g., by the guanidinium-thiocyanate extraction procedure of Chirgwin et al., 1979 *Biochemistry* 18:5294-5299) and cDNA can be prepared using reverse transcriptase (e.g., Moloney MLV reverse transcriptase, available from Gibco/BRL, Bethesda, Md.; or AMV reverse transcriptase, available from Seikagaku America, Inc., St. Petersburg, Fla.). Synthetic oligonucleotide primers for polymerase chain reaction amplification can be designed based upon one of the nucleotide sequences shown in SEQ ID NO:1, SEQ ID NO:2, SEQ ID NO:3, SEQ ID NO:4, SEQ ID NO:5, SEQ ID NO:6, SEQ ID NO:7, SEQ ID NO:8, SEQ ID NO:9, SEQ ID NO:10, SEQ ID NO:11, SEQ ID NO:12 and SEQ ID NO:13. A nucleic acid molecule of the invention can be amplified using cDNA or, alternatively, genomic DNA, as a template and appropriate oligonucleotide primers according to standard PCR amplification techniques. The nucleic acid molecule so amplified can be cloned into an appropriate vector and characterized by DNA sequence analysis. Furthermore, oligonucleotides corresponding to a PKSRP nucleotide sequence can be prepared by standard synthetic techniques, e.g., using an automated DNA synthesizer.

In a preferred embodiment, an isolated nucleic acid molecule of the invention comprises one of the nucleotide sequences shown in SEQ ID NO:14, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24, SEQ ID NO:25 and SEQ ID NO:26. These cDNAs comprise sequences encoding the PKSRLPs (i.e., the "coding region", indicated in Table 1), as well as 5' untranslated sequences and 3' untranslated sequences. It is to be understood that SEQ ID NO:14, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24, SEQ ID NO:25 and SEQ ID NO:26 comprise both coding regions and 5' and 3' untranslated regions. Alternatively, the nucleic acid molecules of the present invention can comprise only the coding region of any of the sequences in SEQ ID NO:14, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24, SEQ ID NO:25 or SEQ ID NO:26 or can contain whole genomic fragments isolated from genomic DNA. A coding region of these sequences is indicated as "ORF position". The present invention also includes PKSRP coding nucleic acids that encode PKSRLPs as described herein. Preferred is a PKSRP coding nucleic acid that encodes a PKSRP selected from the group consisting of, PK-6 (SEQ ID NO:27), PK-7 (SEQ ID NO:28), PK-8 (SEQ ID NO:29), PK-9 (SEQ ID NO:30), CK-1 (SEQ ID NO:31), CK-2 (SEQ

ID NO:32), CK-3 (SEQ ID NO:33), MPK-2 (SEQ ID NO:34), MPK-3 (SEQ ID NO:35), MPK-4 (SEQ ID NO:36), MPK-5 (SEQ ID NO:37), CPK-1 (SEQ ID NO:38) and CPK-2 (SEQ ID NO:39).

Moreover, the nucleic acid molecule of the invention can comprise only a portion of the coding region of one of the sequences in SEQ ID NO:14, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24, SEQ ID NO:25 and SEQ ID NO:26, for example, a fragment which can be used as a probe or primer or a fragment encoding a biologically active portion of a PKSRRP. The nucleotide sequences determined from the cloning of the PKSRRP genes from *P. patens* allow for the generation of probes and primers designed for use in identifying and/or cloning PKSRRP homologs in other cell types and organisms, as well as PKSRRP homologs from other mosses and related species.

Portions of proteins encoded by the PKSRRP nucleic acid molecules of the invention are preferably biologically active portions of one of the PKSRRPs described herein. As used herein, the term "biologically active portion of" a PKSRRP is intended to include a portion, e.g., a domain/motif, of a PKSRRP that participates in a stress tolerance response in a plant, has an activity as set forth in Table 1, or participates in the transcription of a protein involved in a stress tolerance response in a plant. To determine whether a PKSRRP, or a biologically active portion thereof, can participate in transcription of a protein involved in a stress tolerance response in a plant, or whether repression of a PKSRRP results in increased stress tolerance in a plant, a stress analysis of a plant comprising the PKSRRP may be performed. Such analysis methods are well known to those skilled in the art, as detailed in Example 7. More specifically, nucleic acid fragments encoding biologically active portions of a PKSRRP can be prepared by isolating a portion of one of the sequences in SEQ ID NO:27, SEQ ID NO:28, SEQ ID NO:29, SEQ ID NO:30, SEQ ID NO:31, SEQ ID NO:32, SEQ ID NO:33, SEQ ID NO:34, SEQ ID NO:35, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:38 and SEQ ID NO:39, expressing the encoded portion of the PKSRRP or peptide (e.g., by recombinant expression *in vitro*) and assessing the activity of the encoded portion of the PKSRRP or peptide.

Biologically active portions of a PKSRRP are encompassed by the present invention and include peptides comprising amino acid sequences derived from the amino acid sequence of a PKSRRP, e.g., an amino acid sequence of SEQ ID NO:27, SEQ ID NO:28, SEQ ID NO:29, SEQ ID NO:30, SEQ ID NO:31, SEQ ID NO:32, SEQ ID NO:33, SEQ ID NO:34, SEQ ID NO:35, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:38 or SEQ ID NO:39, or the amino acid sequence of a protein homologous to a PKSRRP, which include fewer amino acids than a full length PKSRRP or the full length protein which is homologous to a PKSRRP, and exhibit at least one activity of a PKSRRP. Typically, biologically active portions (e.g., peptides which are, for example, 5, 10, 15, 20, 30, 35, 36, 37, 38, 39, 40, 50, 100 or more amino acids in length) comprise a domain or motif with at least one activity of a PKSRRP. Moreover, other biologically active portions in which other regions of the protein are deleted, can be prepared by recombinant techniques and evaluated for one or more of the activities described herein. Preferably, the biologically active portions of a PKSRRP include one or more selected domains/motifs or portions thereof having biological activity.

The invention also provides PKSRRP chimeric or fusion proteins. As used herein, a PKSRRP "chimeric protein" or "fusion protein" comprises a PKSRRP polypeptide operatively

linked to a non-PKSRRP polypeptide. A PKSRRP polypeptide refers to a polypeptide having an amino acid sequence corresponding to a PKSRRP, whereas a non-PKSRRP polypeptide refers to a polypeptide having an amino acid sequence corresponding to a protein which is not substantially homologous to the PKSRRP, e.g., a protein that is different from the PKSRRP and is derived from the same or a different organism. Within the fusion protein, the term "operatively linked" is intended to indicate that the PKSRRP polypeptide and the non-PKSRRP polypeptide are fused to each other so that both sequences fulfill the proposed function attributed to the sequence used. The non-PKSRRP polypeptide can be fused to the N-terminus or C-terminus of the PKSRRP polypeptide. For example, in one embodiment, the fusion protein is a GST-PKSRRP fusion protein in which the PKSRRP sequences are fused to the C-terminus of the GST sequences. Such fusion proteins can facilitate the purification of recombinant PKSRRPs. In another embodiment, the fusion protein is a PKSRRP containing a heterologous signal sequence at its N-terminus. In certain host cells (e.g., mammalian host cells), expression and/or secretion of a PKSRRP can be increased through use of a heterologous signal sequence.

Preferably, a PKSRRP chimeric or fusion protein of the invention is produced by standard recombinant DNA techniques. For example, DNA fragments coding for the different polypeptide sequences are ligated together in-frame in accordance with conventional techniques, for example by employing blunt-ended or stagger-ended termini for ligation, restriction enzyme digestion to provide for appropriate termini, filling-in of cohesive ends as appropriate, alkaline phosphatase treatment to avoid undesirable joining and enzymatic ligation. In another embodiment, the fusion gene can be synthesized by conventional techniques including automated DNA synthesizers. Alternatively, PCR amplification of gene fragments can be carried out using anchor primers which give rise to complementary overhangs between two consecutive gene fragments which can subsequently be annealed and re-amplified to generate a chimeric gene sequence (see, for example, Current Protocols in Molecular Biology, Eds. Ausubel et al. John Wiley & Sons: 1992). Moreover, many expression vectors are commercially available that already encode a fusion moiety (e.g., a GST polypeptide). A PKSRRP encoding nucleic acid can be cloned into such an expression vector such that the fusion moiety is linked in-frame to the PKSRRP.

In addition to fragments and fusion proteins of the PKSRRPs described herein, the present invention includes homologs and analogs of naturally occurring PKSRRPs and PKSRRP encoding nucleic acids in a plant. "Homologs" are defined herein as two nucleic acids or proteins that have similar, or "homologous", nucleotide or amino acid sequences, respectively. Homologs include allelic variants, orthologs, paralogs, agonists and antagonists of PKSRRPs as defined hereafter. The term "homolog" further encompasses nucleic acid molecules that differ from one of the nucleotide sequences shown in SEQ ID NO:14, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24, SEQ ID NO:25 and SEQ ID NO:26 (and portions thereof) due to degeneracy of the genetic code and thus encode the same PKSRRP as that encoded by the nucleotide sequences shown in SEQ ID NO:14, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24, SEQ ID NO:25 or SEQ ID NO:26. As used herein a "naturally occurring" PKSRRP refers to a PKSRRP amino acid sequence that occurs in nature. Prefer-



ably, a naturally occurring PKSRRP comprises an amino acid sequence selected from the group consisting of SEQ ID NO:27, SEQ ID NO:28, SEQ ID NO:29, SEQ ID NO:30, SEQ ID NO:31, SEQ ID NO:32, SEQ ID NO:33, SEQ ID NO:34, SEQ ID NO:35, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:38 and SEQ ID NO:39.

An agonist of the PKSRRP can retain substantially the same, or a subset, of the biological activities of the PKSRRP. An antagonist of the PKSRRP can inhibit one or more of the activities of the naturally occurring form of the PKSRRP. For example, the PKSRRP antagonist can competitively bind to a downstream or upstream member of the cell membrane component metabolic cascade that includes the PKSRRP, or bind to a PKSRRP that mediates transport of compounds across such membranes, thereby preventing translocation from taking place.

Nucleic acid molecules corresponding to natural allelic variants and analogs, orthologs and paralogs of a PKSRRP cDNA can be isolated based on their identity to the *Physcomitrella patens* PKSRRP nucleic acids described herein using PKSRRP cDNAs, or a portion thereof, as a hybridization probe according to standard hybridization techniques under stringent hybridization conditions. In an alternative embodiment, homologs of the PKSRRP can be identified by screening combinatorial libraries of mutants, e.g., truncation mutants, of the PKSRRP for PKSRRP agonist or antagonist activity. In one embodiment, a variegated library of PKSRRP variants is generated by combinatorial mutagenesis at the nucleic acid level and is encoded by a variegated gene library. A variegated library of PKSRRP variants can be produced by, for example, enzymatically ligating a mixture of synthetic oligonucleotides into gene sequences such that a degenerate set of potential PKSRRP sequences is expressible as individual polypeptides, or alternatively, as a set of larger fusion proteins (e.g., for phage display) containing the set of PKSRRP sequences therein. There are a variety of methods that can be used to produce libraries of potential PKSRRP homologs from a degenerate oligonucleotide sequence. Chemical synthesis of a degenerate gene sequence can be performed in an automatic DNA synthesizer, and the synthetic gene is then ligated into an appropriate expression vector. Use of a degenerate set of genes allows for the provision, in one mixture, of all of the sequences encoding the desired set of potential PKSRRP sequences. Methods for synthesizing degenerate oligonucleotides are known in the art (see, e.g., Narang, S. A., 1983 Tetrahedron 39:3; Itakura et al., 1984 Annu. Rev. Biochem. 53:323; Itakura et al., 1984 Science 198:1056; Ike et al., 1983 Nucleic Acid Res. 11:477).

In addition, libraries of fragments of the PKSRRP coding regions can be used to generate a variegated population of PKSRRP fragments for screening and subsequent selection of homologs of a PKSRRP. In one embodiment, a library of coding sequence fragments can be generated by treating a double stranded PCR fragment of a PKSRRP coding sequence with a nuclease under conditions wherein nicking occurs only about once per molecule, denaturing the double stranded DNA, renaturing the DNA to form double stranded DNA, which can include sense/antisense pairs from different nicked products, removing single stranded portions from reformed duplexes by treatment with S I nuclease, and ligating the resulting fragment library into an expression vector. By this method, an expression library can be derived which encodes N-terminal, C-terminal and internal fragments of various sizes of the PKSRRP.

Several techniques are known in the art for screening gene products of combinatorial libraries made by point mutations or truncation, and for screening cDNA libraries for gene

products having a selected property. Such techniques are adaptable for rapid screening of the gene libraries generated by the combinatorial mutagenesis of PKSRRP homologs. The most widely used techniques, which are amenable to high through-put analysis, for screening large gene libraries typically include cloning the gene library into replicable expression vectors, transforming appropriate cells with the resulting library of vectors, and expressing the combinatorial genes under conditions in which detection of a desired activity facilitates isolation of the vector encoding the gene whose product was detected. Recursive ensemble mutagenesis (REM), a new technique that enhances the frequency of functional mutants in the libraries, can be used in combination with the screening assays to identify PKSRRP homologs (Arkin and Yourvan, 1992 PNAS 89:7811-7815; Delgrave et al., 1993 Protein Engineering 6(3):327-331). In another embodiment, cell based assays can be exploited to analyze a variegated PKSRRP library, using methods well known in the art. The present invention further provides a method of identifying a novel PKSRRP, comprising (a) raising a specific antibody response to a PKSRRP, or a fragment thereof, as described herein; (b) screening putative PKSRRP material with the antibody, wherein specific binding of the antibody to the material indicates the presence of a potentially novel PKSRRP; and (c) analyzing the bound material in comparison to known PKSRRP, to determine its novelty.

To determine the percent homology of two amino acid sequences (e.g., one of the sequences of SEQ ID NO:27, SEQ ID NO:28, SEQ ID NO:29, SEQ ID NO:30, SEQ ID NO:31, SEQ ID NO:32, SEQ ID NO:33, SEQ ID NO:34, SEQ ID NO:35, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:38 and SEQ ID NO:39 and a mutant form thereof), the sequences are aligned for optimal comparison purposes (e.g., gaps can be introduced in the sequence of one protein or nucleic acid for optimal alignment with the other protein or nucleic acid). The amino acid residues at corresponding amino acid positions or nucleotide positions are then compared. When a position in one sequence (e.g., one of the sequences of SEQ ID NO:27, SEQ ID NO:28, SEQ ID NO:29, SEQ ID NO:30, SEQ ID NO:31, SEQ ID NO:32, SEQ ID NO:33, SEQ ID NO:34, SEQ ID NO:35, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:38 and SEQ ID NO:39) is occupied by the same amino acid residue as the corresponding position in the other sequence (e.g., a mutant form of the sequence selected from the polypeptide of SEQ ID NO:27, SEQ ID NO:28, SEQ ID NO:29, SEQ ID NO:30, SEQ ID NO:31, SEQ ID NO:32, SEQ ID NO:33, SEQ ID NO:34, SEQ ID NO:35, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:38 and SEQ ID NO:39), then the molecules are homologous at that position (i.e., as used herein amino acid or nucleic acid "homology" is equivalent to amino acid or nucleic acid "identity"). The same type of comparison can be made between two nucleic acid sequences.

The percent homology between the two sequences is a function of the number of identical positions shared by the sequences (i.e., % homology = numbers of identical positions / total numbers of positions x 100). Preferably, the amino acid sequences included in the present invention are at least about 50-60%, preferably at least about 60-70%, and more preferably at least about 70-80%, 80-90%, 90-95%, and most preferably at least about 96%, 97%, 98%, 99% or more homologous to an entire amino acid sequence shown in SEQ ID NO:27, SEQ ID NO:28, SEQ ID NO:29, SEQ ID NO:30, SEQ ID NO:31, SEQ ID NO:32, SEQ ID NO:33, SEQ ID NO:34, SEQ ID NO:35, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:38 or SEQ ID NO:39. In yet another embodiment, at least about 50-60%, preferably at least about

60-70%, and more preferably at least about 70-80%, 80-90%, 90-95%, and most preferably at least about 96%, 97%, 98%, 99% or more homologous to an entire amino acid sequence encoded by a nucleic acid sequence shown in SEQ ID NO:14, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24, SEQ ID NO:25 or SEQ ID NO:26. In other embodiments, the preferable length of sequence comparison for proteins is at least 15 amino acid residues, more preferably at least 25 amino acid residues, and most preferably at least 35 amino acid residues.

In another preferred embodiment, an isolated nucleic acid molecule of the invention comprises a nucleotide sequence which is at least about 50-60%, preferably at least about 60-70%, more preferably at least about 70-80%, 80-90%, 90-95%, and even more preferably at least about 95%, 96%, 97%, 98%, 99% or more homologous to a nucleotide sequence shown in SEQ ID NO:14, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24, SEQ ID NO:25 or SEQ ID NO:26, or a portion thereof. The preferable length of sequence comparison for nucleic acids is at least 75 nucleotides, more preferably at least 100 nucleotides and most preferably the entire length of the coding region.

It is also preferable that the homologous nucleic acid molecule of the invention encodes a protein or portion thereof which includes an amino acid sequence which is sufficiently homologous to an amino acid sequence of SEQ ID NO:27, SEQ ID NO:28, SEQ ID NO:29, SEQ ID NO:30, SEQ ID NO:31, SEQ ID NO:32, SEQ ID NO:33, SEQ ID NO:34, SEQ ID NO:35, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:38 or SEQ ID NO:39 such that the protein or portion thereof maintains the same or a similar function as the amino acid sequence to which it is compared. Functions of the PKSRP amino acid sequences of the present invention include the ability to participate in a stress tolerance response in a plant, or more particularly, to participate in the transcription of a protein involved in a stress tolerance response in a *Physcomitrella patens* plant. Examples of such activities are described in Table 1.

In addition to the above described methods, a determination of the percent homology between two sequences can be accomplished using a mathematical algorithm. A preferred, non-limiting example of a mathematical algorithm utilized for the comparison of two sequences is the algorithm of Karlin and Altschul (1990 Proc. Natl. Acad. Sci. USA 90:5873-5877). Such an algorithm is incorporated into the NBLAST and XBLAST programs of Altschul, et al. (1990 J. Mol. Biol. 215:403-410).

BLAST nucleic acid searches can be performed with the NBLAST program, score=100, wordlength=12 to obtain nucleic acid sequences homologous to the PKSRP nucleic acid molecules of the invention. Additionally, BLAST protein searches can be performed with the XBLAST program, score=50, wordlength=3 to obtain amino acid sequences homologous to PKSRPs of the present invention. To obtain gapped alignments for comparison purposes, Gapped BLAST can be utilized as described in Altschul et al. (1997 Nucleic Acids Res. 25:3389-3402). When utilizing BLAST and Gapped BLAST programs, the default parameters of the respective programs (e.g., XBLAST and NBLAST) can be used. Another preferred non-limiting example of a mathematical algorithm utilized for the comparison of sequences is the algorithm of Myers and Miller (CABIOS 1989). Such an algorithm is incorporated into the ALIGN program (version 2.0) that is part of the GCG sequence alignment software

package. When utilizing the ALIGN program for comparing amino acid sequences, a PAM120 weight residue table, a gap length penalty of 12 and a gap penalty of 4 can be used to obtain amino acid sequences homologous to the PKSRPs of the present invention. To obtain gapped alignments for comparison purposes, Gapped BLAST can be utilized as described in Altschul et al. (1997 Nucleic Acids Res. 25:3389-3402). When utilizing BLAST and Gapped BLAST programs, the default parameters of the respective programs (e.g., XBLAST and NBLAST) can be used. Another preferred non-limiting example of a mathematical algorithm utilized for the comparison of sequences is the algorithm of Myers and Miller (CABIOS 1989). Such an algorithm is incorporated into the ALIGN program (version 2.0) that is part of the GCG sequence alignment software package. When utilizing the ALIGN program for comparing amino acid sequences, a PAM120 weight residue table, a gap length penalty of 12 and a gap penalty of 4 can be used.

Finally, homology between nucleic acid sequences can also be determined using hybridization techniques known to those of skill in the art. Accordingly, an isolated nucleic acid molecule of the invention comprises a nucleotide sequence which hybridizes, e.g., hybridizes under stringent conditions, to one of the nucleotide sequences shown in SEQ ID NO:14, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24, SEQ ID NO:25 and SEQ ID NO:26, or a portion thereof. More particularly, an isolated nucleic acid molecule of the invention is at least 15 nucleotides in length and hybridizes under stringent conditions to the nucleic acid molecule comprising a nucleotide sequence of SEQ ID NO:14, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24, SEQ ID NO:25 or SEQ ID NO:26. In other embodiments, the nucleic acid is at least 30, 50, 100, 250 or more nucleotides in length.

As used herein, the term "hybridizes under stringent conditions" is intended to describe conditions for hybridization and washing under which nucleotide sequences at least 60% homologous to each other typically remain hybridized to each other. Preferably, the conditions are such that sequences at least about 65%, more preferably at least about 70%, and even more preferably at least about 75% or more homologous to each other typically remain hybridized to each other. Such stringent conditions are known to those skilled in the art and can be found in *Current Protocols in Molecular Biology*, 6.3.1-6.3.6, John Wiley & Sons, N.Y. (1989). A preferred, non-limiting example of stringent hybridization conditions are hybridization in 6x sodium chloride/sodium citrate (SSC) at about 45° C., followed by one or more washes in 0.2xSSC, 0.1% SDS at 50-65° C. Preferably, an isolated nucleic acid molecule of the invention that hybridizes under stringent conditions to a sequence of SEQ ID NO:14, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24, SEQ ID NO:25 or SEQ ID NO:26 corresponds to a naturally occurring nucleic acid molecule. As used herein, a "naturally occurring" nucleic acid molecule refers to an RNA or DNA molecule having a nucleotide sequence that occurs in nature (e.g., encodes a natural protein). In one embodiment, the nucleic acid encodes a naturally occurring *Physcomitrella patens* PKSRP.

Using the above-described methods, and others known to those of skill in the art, one of ordinary skill in the art can isolate homologs of the PKSRPs comprising amino acid sequences shown in SEQ ID NO:27, SEQ ID NO:28, SEQ ID

NO:29, SEQ ID NO:30, SEQ ID NO:31, SEQ ID NO:32, SEQ ID NO:33, SEQ ID NO:34, SEQ ID NO:35, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:38 or SEQ ID NO:39. One subset of these homologs are allelic variants. As used herein, the term “allelic variant” refers to a nucleotide sequence containing polymorphisms that lead to changes in the amino acid sequences of a PKSRP and that exist within a natural population (e.g., a plant species or variety). Such natural allelic variations can typically result in 1-5% variance in a PKSRP nucleic acid. Allelic variants can be identified by sequencing the nucleic acid sequence of interest in a number of different plants, which can be readily carried out by using hybridization probes to identify the same PKSRP genetic locus in those plants. Any and all such nucleic acid variations and resulting amino acid polymorphisms or variations in a PKSRP that are the result of natural allelic variation and that do not alter the functional activity of a PKSRP, are intended to be within the scope of the invention.

Moreover, nucleic acid molecules encoding PKSRPs from the same or other species such as PKSRP analogs, orthologs and paralogs, are intended to be within the scope of the present invention. As used herein, the term “analog” refers to two nucleic acids that have the same or similar function, but that have evolved separately in unrelated organisms. As used herein, the term “orthologs” refers to two nucleic acids from different species, but that have evolved from a common ancestral gene by speciation. Normally, orthologs encode proteins having the same or similar functions. As also used herein, the term “paralogs” refers to two nucleic acids that are related by duplication within a genome. Paralogs usually have different functions, but these functions may be related (Tatusov, R. L. et al. 1997 Science 278(5338):631-637). Analog, orthologs and paralogs of a naturally occurring PKSRP can differ from the naturally occurring PKSRP by post-translational modifications, by amino acid sequence differences, or by both. Post-translational modifications include in vivo and in vitro chemical derivatization of polypeptides, e.g., acetylation, carboxylation, phosphorylation, or glycosylation, and such modifications may occur during polypeptide synthesis or processing or following treatment with isolated modifying enzymes. In particular, orthologs of the invention will generally exhibit at least 80-85%, more preferably 90%, and most preferably 95%, 96%, 97%, 98% or even 99% identity or homology with all or part of a naturally occurring PKSRP amino acid sequence and will exhibit a function similar to a PKSRP. Orthologs of the present invention are also preferably capable of participating in the stress response in plants. In one embodiment, the PKSRP orthologs maintain the ability to participate in the metabolism of compounds necessary for the construction of cellular membranes in *Physcomitrella patens*, or in the transport of molecules across these membranes.

In addition to naturally-occurring variants of a PKSRP sequence that may exist in the population, the skilled artisan will further appreciate that changes can be introduced by mutation into a nucleotide sequence of SEQ ID NO:14, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24, SEQ ID NO:25 or SEQ ID NO:26, thereby leading to changes in the amino acid sequence of the encoded PKSRP, without altering the functional ability of the PKSRP. For example, nucleotide substitutions leading to amino acid substitutions at “non-essential” amino acid residues can be made in a sequence of SEQ ID NO:14, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24,

SEQ ID NO:25 or SEQ ID NO:26. A “non-essential” amino acid residue is a residue that can be altered from the wild-type sequence of one of the PKSRPs without altering the activity of said PKSRP, whereas an “essential” amino acid residue is required for PKSRP activity. Other amino acid residues, however, (e.g., those that are not conserved or only semi-conserved in the domain having PKSRP activity) may not be essential for activity and thus are likely to be amenable to alteration without altering PKSRP activity.

Accordingly, another aspect of the invention pertains to nucleic acid molecules encoding PKSRPs that contain changes in amino acid residues that are not essential for PKSRP activity. Such PKSRPs differ in amino acid sequence from a sequence contained in SEQ ID NO:27, SEQ ID NO:28, SEQ ID NO:29, SEQ ID NO:30, SEQ ID NO:31, SEQ ID NO:32, SEQ ID NO:33, SEQ ID NO:34, SEQ ID NO:35, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:38 or SEQ ID NO:39, yet retain at least one of the PKSRP activities described herein. In one embodiment, the isolated nucleic acid molecule comprises a nucleotide sequence encoding a protein, wherein the protein comprises an amino acid sequence at least about 50% homologous to an amino acid sequence of SEQ ID NO:27, SEQ ID NO:28, SEQ ID NO:29, SEQ ID NO:30, SEQ ID NO:31, SEQ ID NO:32, SEQ ID NO:33, SEQ ID NO:34, SEQ ID NO:35, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:38 and SEQ ID NO:39. Preferably, the protein encoded by the nucleic acid molecule is at least about 50-60% homologous to one of the sequences of SEQ ID NO:27, SEQ ID NO:28, SEQ ID NO:29, SEQ ID NO:30, SEQ ID NO:31, SEQ ID NO:32, SEQ ID NO:33, SEQ ID NO:34, SEQ ID NO:35, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:38 and SEQ ID NO:39, more preferably at least about 60-70% homologous to one of the sequences of SEQ ID NO:27, SEQ ID NO:28, SEQ ID NO:29, SEQ ID NO:30, SEQ ID NO:31, SEQ ID NO:32, SEQ ID NO:33, SEQ ID NO:34, SEQ ID NO:35, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:38 and SEQ ID NO:39, even more preferably at least about 70-80%, 80-90%, 90-95% homologous to one of the sequences of SEQ ID NO:27, SEQ ID NO:28, SEQ ID NO:29, SEQ ID NO:30, SEQ ID NO:31, SEQ ID NO:32, SEQ ID NO:33, SEQ ID NO:34, SEQ ID NO:35, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:38 and SEQ ID NO:39, and most preferably at least about 96%, 97%, 98%, or 99% homologous to one of the sequences of SEQ ID NO:27, SEQ ID NO:28, SEQ ID NO:29, SEQ ID NO:30, SEQ ID NO:31, SEQ ID NO:32, SEQ ID NO:33, SEQ ID NO:34, SEQ ID NO:35, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:38 and SEQ ID NO:39. The preferred PKSRP homologs of the present invention are preferably capable of participating in the a stress tolerance response in a plant, or more particularly, participating in the transcription of a protein involved in a stress tolerance response in a *Physcomitrella patens* plant, or have one or more activities set forth in Table 1.

An isolated nucleic acid molecule encoding a PKSRP homologous to a protein sequence of SEQ ID NO:27, SEQ ID NO:28, SEQ ID NO:29, SEQ ID NO:30, SEQ ID NO:31, SEQ ID NO:32, SEQ ID NO:33, SEQ ID NO:34, SEQ ID NO:35, SEQ ID NO:36, SEQ ID NO:37, SEQ ID NO:38 or SEQ ID NO:39 can be created by introducing one or more nucleotide substitutions, additions or deletions into a nucleotide sequence of SEQ ID NO:14, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24, SEQ ID NO:25 or SEQ ID NO:26 such that one or more amino acid substitutions, additions or deletions are introduced into the encoded protein. Mutations

can be introduced into one of the sequences of SEQ ID NO:14, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24, SEQ ID NO:25 and SEQ ID NO:26 by standard techniques, such as site-directed mutagenesis and PCR-mediated mutagenesis. Preferably, conservative amino acid substitutions are made at one or more predicted non-essential amino acid residues. A "conservative amino acid substitution" is one in which the amino acid residue is replaced with an amino acid residue having a similar side chain.

Families of amino acid residues having similar side chains have been defined in the art. These families include amino acids with basic side chains (e.g., lysine, arginine, histidine), acidic side chains (e.g., aspartic acid, glutamic acid), uncharged polar side chains (e.g., glycine, asparagine, glutamine, serine, threonine, tyrosine, cysteine), nonpolar side chains (e.g., alanine, valine, leucine, isoleucine, proline, phenylalanine, methionine, tryptophan), beta-branched side chains (e.g., threonine, valine, isoleucine) and aromatic side chains (e.g., tyrosine, phenylalanine, tryptophan, histidine). Thus, a predicted nonessential amino acid residue in a PKSRP is preferably replaced with another amino acid residue from the same side chain family. Alternatively, in another embodiment, mutations can be introduced randomly along all or part of a PKSRP coding sequence, such as by saturation mutagenesis, and the resultant mutants can be screened for a PKSRP activity described herein to identify mutants that retain PKSRP activity. Following mutagenesis of one of the sequences of SEQ ID NO:14, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24, SEQ ID NO:25 and SEQ ID NO:26, the encoded protein can be expressed recombinantly and the activity of the protein can be determined by analyzing the stress tolerance of a plant expressing the protein as described in Example 7.

In addition to the nucleic acid molecules encoding the PKSRPs described above, another aspect of the invention pertains to isolated nucleic acid molecules that are antisense thereto. An "antisense" nucleic acid comprises a nucleotide sequence that is complementary to a "sense" nucleic acid encoding a protein, e.g., complementary to the coding strand of a double-stranded cDNA molecule or complementary to an mRNA sequence. Accordingly, an antisense nucleic acid can hydrogen bond to a sense nucleic acid. The antisense nucleic acid can be complementary to an entire PKSRP coding strand, or to only a portion thereof. In one embodiment, an antisense nucleic acid molecule is antisense to a "coding region" of the coding strand of a nucleotide sequence encoding a PKSRP. The term "coding region" refers to the region of the nucleotide sequence comprising codons that are translated into amino acid residues (e.g., the entire coding region of . . . comprises nucleotides 1 to . . .). In another embodiment, the antisense nucleic acid molecule is antisense to a "noncoding region" of the coding strand of a nucleotide sequence encoding a PKSRP. The term "noncoding region" refers to 5' and 3' sequences that flank the coding region that are not translated into amino acids (i.e., also referred to as 5' and 3' untranslated regions).

In a preferred embodiment, an isolated, nucleic acid molecule of the invention comprises a nucleic acid molecule which is a complement of one of the nucleotide sequences shown in SEQ ID NO:14, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24, SEQ ID NO:25 and SEQ ID NO:26, or a

portion thereof. A nucleic acid molecule that is complementary to one of the nucleotide sequences shown in SEQ ID NO:14, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24, SEQ ID NO:25 and SEQ ID NO:26 is one which is sufficiently complementary to one of the nucleotide sequences shown in SEQ ID NO:14, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24, SEQ ID NO:25 and SEQ ID NO:26 such that it can hybridize to one of the nucleotide sequences shown in SEQ ID NO:14, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24, SEQ ID NO:25 and SEQ ID NO:26, thereby forming a stable duplex.

Given the coding strand sequences encoding the PKSRPs disclosed herein (e.g., the sequences set forth in SEQ ID NO:14, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24, SEQ ID NO:25 and SEQ ID NO:26), antisense nucleic acids of the invention can be designed according to the rules of Watson and Crick base pairing. The antisense nucleic acid molecule can be complementary to the entire coding region of PKSRP mRNA, but more preferably is an oligonucleotide which is antisense to only a portion of the coding or noncoding region of PKSRP mRNA. For example, the antisense oligonucleotide can be complementary to the region surrounding the translation start site of PKSRP mRNA. An antisense oligonucleotide can be, for example, about 5, 10, 15, 20, 25, 30, 35, 40, 45 or 50 nucleotides in length.

An antisense nucleic acid of the invention can be constructed using chemical synthesis and enzymatic ligation reactions using procedures known in the art. For example, an antisense nucleic acid (e.g., an antisense oligonucleotide) can be chemically synthesized using naturally occurring nucleotides or variously modified nucleotides designed to increase the biological stability of the molecules or to increase the physical stability of the duplex formed between the antisense and sense nucleic acids, e.g., phosphorothioate derivatives and acridine substituted nucleotides can be used. Examples of modified nucleotides which can be used to generate the antisense nucleic acid include 5-fluorouracil, 5-bromouracil, 5-chlorouracil, 5-iodouracil, hypoxanthine, xanthine, 4-acetylcytosine, 5-(carboxyhydroxymethyl) uracil, 5-carboxymethylaminomethyl-2-thiouridine, 5-carboxymethylaminomethyluracil, dihydrouracil, beta-D-galactosylqueosine, inosine, N6-isopentenyladenine, 1-methylguanine, 1-methylinosine, 2,2-dimethylguanine, 2-methyladenine, 2-methylguanine, 3-methylcytosine, 5-methylcytosine, N6-adenine, 7-methylguanine, 5-methylaminomethyluracil, 5-methoxyaminomethyl-2-thiouracil, beta-D-mannosylqueosine, 5'-methoxycarbonylmethyluracil, 5-methoxyuracil, 2-methylthio-N6-isopentenyladenine, uracil-5-oxyacetic acid (v), wybutoxosine, pseudouracil, queosine, 2-thiocytosine, 5-methyl-2-thiouracil, 2-thiouracil, 4-thiouracil, 5-methyluracil, uracil-5-oxyacetic acid methylester, uracil-5-oxyacetic acid (v), 5-methyl-2-thiouracil, 3-(3-amino-3-N-2-carboxypropyl)uracil, (acp3)w, and 2,6-diaminopurine. Alternatively, the antisense nucleic acid can be produced biologically using an expression vector into which a nucleic acid has been subcloned in an antisense orientation (i.e., RNA transcribed from the inserted nucleic acid will be of an antisense orientation to a target nucleic acid of interest, described further in the following subsection).

The antisense nucleic acid molecules of the invention are typically administered to a cell or generated in situ such that they hybridize with or bind to cellular mRNA and/or genomic DNA encoding a PKSRP to thereby inhibit expression of the protein, e.g., by inhibiting transcription and/or translation. The hybridization can be by conventional nucleotide complementarity to form a stable duplex, or, for example, in the case of an antisense nucleic acid molecule which binds to DNA duplexes, through specific interactions in the major groove of the double helix. The antisense molecule can be modified such that it specifically binds to a receptor or an antigen expressed on a selected cell surface, e.g., by linking the antisense nucleic acid molecule to a peptide or an antibody which binds to a cell surface receptor or antigen. The antisense nucleic acid molecule can also be delivered to cells using the vectors described herein. To achieve sufficient intracellular concentrations of the antisense molecules, vector constructs in which the antisense nucleic acid molecule is placed under the control of a strong prokaryotic, viral, or eukaryotic (including plant) promoter are preferred.

In yet another embodiment, the antisense nucleic acid molecule of the invention is an  $\alpha$ -anomeric nucleic acid molecule. An  $\alpha$ -anomeric nucleic acid molecule forms specific double-stranded hybrids with complementary RNA in which, contrary to the usual  $\beta$ -units, the strands run parallel to each other (Gaultier et al., 1987 Nucleic Acids. Res. 15:6625-6641). The antisense nucleic acid molecule can also comprise a 2'-O-methylribonucleotide (Inoue et al., 1987 Nucleic Acids Res. 15:6131-6148) or a chimeric RNA-DNA analogue (Inoue et al., 1987 FEBS Lett. 215:327-330).

In still another embodiment, an antisense nucleic acid of the invention is a ribozyme. Ribozymes are catalytic RNA molecules with ribonuclease activity which are capable of cleaving a single-stranded nucleic acid, such as an mRNA, to which they have a complementary region. Thus, ribozymes (e.g., hammerhead ribozymes described in Haselhoff and Gerlach, 1988 Nature 334:585-591) can be used to catalytically cleave PKSRP mRNA transcripts to thereby inhibit translation of PKSRP mRNA. A ribozyme having specificity for a PKSRP-encoding nucleic acid can be designed based upon the nucleotide sequence of a PKSRP cDNA, as disclosed herein (i.e., SEQ ID NO:14, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24, SEQ ID NO:25 or SEQ ID NO:26) or on the basis of a heterologous sequence to be isolated according to methods taught in this invention. For example, a derivative of a *Tetrahymena* L-19 IVS RNA can be constructed in which the nucleotide sequence of the active site is complementary to the nucleotide sequence to be cleaved in a PKSRP-encoding mRNA. See, e.g., Cecil et al. U.S. Pat. No. 4,987,071 and Cecil et al. U.S. Pat. No. 5,116,742. Alternatively, PKSRP mRNA can be used to select a catalytic RNA having a specific ribonuclease activity from a pool of RNA molecules. See, e.g., Bartel, D. and Szostak, J. W., 1993 Science 261:1411-1418.

Alternatively, PKSRP gene expression can be inhibited by targeting nucleotide sequences complementary to the regulatory region of a PKSRP nucleotide sequence (e.g., a PKSRP promoter and/or enhancer) to form triple helical structures that prevent transcription of a PKSRP gene in target cells. See generally, Helene, C., 1991 Anticancer Drug Des. 6(6):569-84; Helene, C. et al., 1992 Ann. N.Y. Acad. Sci. 660:27-36; and Maher, L. J., 1992 Bioassays 14(12):807-15. 109801 In addition to the PKSRP nucleic acids and proteins described above, the present invention encompasses these nucleic acids and proteins attached to a moiety. These moieties include, but

are not limited to, detection moieties, hybridization moieties, purification moieties, delivery moieties, reaction moieties, binding moieties, and the like. A typical group of nucleic acids having moieties attached are probes and primers. The probes and primers typically comprise a substantially isolated oligonucleotide. The oligonucleotide typically comprises a region of nucleotide sequence that hybridizes under stringent conditions to at least about 12, preferably about 25, more preferably about 40, 50 or 75 consecutive nucleotides of a sense strand of one of the sequences set forth in SEQ ID NO:14, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24, SEQ ID NO:25 and SEQ ID NO:26, an anti-sense sequence of one of the sequences set forth in SEQ ID NO:14, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24, SEQ ID NO:25 and SEQ ID NO:26, or naturally occurring mutants thereof. Primers based on a nucleotide sequence of SEQ ID NO:14, SEQ ID NO:15, SEQ ID NO:16, SEQ ID NO:17, SEQ ID NO:18, SEQ ID NO:19, SEQ ID NO:20, SEQ ID NO:21, SEQ ID NO:22, SEQ ID NO:23, SEQ ID NO:24, SEQ ID NO:25 or SEQ ID NO:26 can be used in PCR reactions to clone PKSRP homologs. Probes based on the PKSRP nucleotide sequences can be used to detect transcripts or genomic sequences encoding the same or homologous proteins. In preferred embodiments, the probe further comprises a label group attached thereto, e.g. the label group can be a radioisotope, a fluorescent compound, an enzyme, or an enzyme co-factor. Such probes can be used as a part of a genomic marker test kit for identifying cells which express a PKSRP, such as by measuring a level of a PKSRP-encoding nucleic acid, in a sample of cells, e.g., detecting PKSRP mRNA levels or determining whether a genomic PKSRP gene has been mutated or deleted.

In particular, a useful method to ascertain the level of transcription of the gene (an indicator of the amount of mRNA available for translation to the gene product) is to perform a Northern blot (for reference see, for example, Ausubel et al., 1988 Current Protocols in Molecular Biology, Wiley: N.Y.). This information at least partially demonstrates the degree of transcription of the transformed gene. Total cellular RNA can be prepared from cells, tissues or organs by several methods, all well-known in the art, such as that described in Bormann, E. R. et al., 1992 Mol. Microbiol. 6:317-326. To assess the presence or relative quantity of protein translated from this mRNA, standard techniques, such as a Western blot, may be employed. These techniques are well known to one of ordinary skill in the art. (See, for example, Ausubel et al., 1988 Current Protocols in Molecular Biology, Wiley: N.Y.).

The invention further provides an isolated recombinant expression vector comprising a PKSRP nucleic acid as described above, wherein expression of the vector in a host cell results in increased tolerance to environmental stress as compared to a wild type variety of the host cell. As used herein, the term "vector" refers to a nucleic acid molecule capable of transporting another nucleic acid to which it has been linked. One type of vector is a "plasmid", which refers to a circular double stranded DNA loop into which additional DNA segments can be ligated. Another type of vector is a viral vector, wherein additional DNA segments can be ligated into the viral genome. Certain vectors are capable of autonomous replication in a host cell into which they are introduced (e.g., bacterial vectors having a bacterial origin of replication and episomal mammalian vectors). Other vectors (e.g., non-episomal mammalian vectors) are integrated into the genome of

a host cell upon introduction into the host cell, and thereby are replicated along with the host genome. Moreover, certain vectors are capable of directing the expression of genes to which they are operatively linked. Such vectors are referred to herein as "expression vectors". In general, expression vectors of utility in recombinant DNA techniques are often in the form of plasmids. In the present specification, "plasmid" and "vector" can be used interchangeably as the plasmid is the most commonly used form of vector. However, the invention is intended to include such other forms of expression vectors, such as viral vectors (e.g., replication defective retroviruses, adenoviruses and adeno-associated viruses), which serve equivalent functions.

The recombinant expression vectors of the invention comprise a nucleic acid of the invention in a form suitable for expression of the nucleic acid in a host cell, which means that the recombinant expression vectors include one or more regulatory sequences, selected on the basis of the host cells to be used for expression, which is operatively linked to the nucleic acid sequence to be expressed. Within a recombinant expression vector, "operably linked" is intended to mean that the nucleotide sequence of interest is linked to the regulatory sequence(s) in a manner which allows for expression of the nucleotide sequence (e.g., in an in vitro transcription/translation system or in a host cell when the vector is introduced into the host cell). The term "regulatory sequence" is intended to include promoters, enhancers and other expression control elements (e.g., polyadenylation signals). Such regulatory sequences are described, for example, in Goeddel, *Gene Expression Technology: Methods in Enzymology* 185, Academic Press, San Diego, Calif. (1990) or see: Gruber and Crosby, in: *Methods in Plant Molecular Biology and Biotechnology*, eds. Glick and Thompson, Chapter 7, 89-108, CRC Press: Boca Raton, Fla., including the references therein. Regulatory sequences include those that direct constitutive expression of a nucleotide sequence in many types of host cells and those that direct expression of the nucleotide sequence only in certain host cells or under certain conditions. It will be appreciated by those skilled in the art that the design of the expression vector can depend on such factors as the choice of the host cell to be transformed, the level of expression of protein desired, etc. The expression vectors of the invention can be introduced into host cells to thereby produce proteins or peptides, including fusion proteins or peptides, encoded by nucleic acids as described herein (e.g., PKSRPs, mutant forms of PKSRPs, fusion proteins, etc.).

The recombinant expression vectors of the invention can be designed for expression of PKSRPs in prokaryotic or eukaryotic cells. For example, PKSRP genes can be expressed in bacterial cells such as *C. glutamicum*, insect cells (using baculovirus expression vectors), yeast and other fungal cells (see Romanos, M. A. et al., 1992 *Foreign gene expression in yeast: a review*, *Yeast* 8:423-488; van den Handel, C. A. M. J. J. et al., 1991 *Heterologous gene expression in filamentous fungi*, in: *More Gene Manipulations in Fungi*, J. W. Bennet & L. L. Lasure, eds., p. 396-428: Academic Press: San Diego; and van den Hondel, C. A. M. J. J. & Punt, P. J., 1991 *Gene transfer systems and vector development for filamentous fungi*, in: *Applied Molecular Genetics of Fungi*, Peberdy, J. F. et al., eds., p. 1-28, Cambridge University Press: Cambridge), algae (Falciatore et al., 1999 *Marine Biotechnology* 1(3):239-251), ciliates of the types: *Holotrichia*, *Peritrichia*, *Spirotrichia*, *Suctorina*, *Tetrahymena*, *Paramecium*, *Colpidium*, *Glaucocoma*, *Platyophrya*, *Potomacus*, *Pseudocohnilembus*, *Euplotes*, *Engelmanniella*, and *Stylonychia*, especially of the genus *Stylonychia lemnae* with vectors following a transformation method as described in WO 98/01572 and multicel-

ular plant cells (see Schmidt, R. and Willmitzer, L., 1988 *High efficiency Agrobacterium tumefaciens-mediated transformation of Arabidopsis thaliana leaf and cotyledon explants*, *Plant Cell Rep.* 583-586); *Plant Molecular Biology and Biotechnology*, C Press, Boca Raton, Fla., chapter 6/7, S.71-119 (1993); F. F. White, B. Jenes et al, *Techniques for Gene Transfer*, in: *Transgenic Plants*, Vol. 1, Engineering and Utilization, eds. Kung and R. Wu, 128-43, Academic Press: 1993; Pottykus, 1991 *Annu. Rev. Plant Physiol. Plant Molec. Biol.* 42:205-225 and references cited therein) or mammalian cells. Suitable host cells are discussed further in Goeddel, *Gene Expression Technology: Methods in Enzymology* 185, Academic Press: San Diego, Calif. (1990). Alternatively, the recombinant expression vector can be transcribed and translated in vitro, for example using T7 promoter regulatory sequences and T7 polymerase.

Expression of proteins in prokaryotes is most often carried out with vectors containing constitutive or inducible promoters directing the expression of either fusion or non-fusion proteins. Fusion vectors add a number of amino acids to a protein encoded therein, usually to the amino terminus of the recombinant protein but also to the C-terminus or fused within suitable regions in the proteins. Such fusion vectors typically serve three purposes: 1) to increase expression of a recombinant protein; 2) to increase the solubility of a recombinant protein; and 3) to aid in the purification of a recombinant protein by acting as a ligand in affinity purification. Often, in fusion expression vectors, a proteolytic cleavage site is introduced at the junction of the fusion moiety and the recombinant protein to enable separation of the recombinant protein from the fusion moiety subsequent to purification of the fusion protein. Such enzymes, and their cognate recognition sequences, include Factor Xa, thrombin and enterokinase.

Typical fusion expression vectors include pGEX (Pharmacia Biotech Inc; Smith, D. B. and Johnson, K. S., 1988 *Gene* 67:31-40), pMAL (New England Biolabs, Beverly, Mass.) and pRIT5 (Pharmacia, Piscataway, N.J.) which fuse glutathione S-transferase (GST), maltose E binding protein, or protein A, respectively, to the target recombinant protein. In one embodiment, the coding sequence of the PKSRP is cloned into a pGEX expression vector to create a vector encoding a fusion protein comprising, from the N-terminus to the C-terminus, GST-thrombin cleavage site-X protein. The fusion protein can be purified by affinity chromatography using glutathione-agarose resin. Recombinant PKSRP unfused to GST can be recovered by cleavage of the fusion protein with thrombin.

Examples of suitable inducible non-fusion *E. coli* expression vectors include pTrc (Amann et al., 1988 *Gene* 69:301-315) and pET 11d (Studier et al., *Gene Expression Technology: Methods in Enzymology* 185, Academic Press, San Diego, Calif. (1990) 60-89). Target gene expression from the pTrc vector relies on host RNA polymerase transcription from a hybrid trp-lac fusion promoter. Target gene expression from the pET 11d vector relies on transcription from a T7 gn10-lac fusion promoter mediated by a co-expressed viral RNA polymerase (T7 gn1). This viral polymerase is supplied by host strains BL21(DE3) or HMS174(DE3) from a resident  $\lambda$  prophage harboring a T7 gn1 gene under the transcriptional control of the lacUV 5 promoter.

One strategy to maximize recombinant protein expression is to express the protein in a host bacteria with an impaired capacity to proteolytically cleave the recombinant protein (Gottesman, S., *Gene Expression Technology: Methods in Enzymology* 185, Academic Press, San Diego, Calif. (1990) 119-128). Another strategy is to alter the sequence of the

nucleic acid to be inserted into an expression vector so that the individual codons for each amino acid are those preferentially utilized in the bacterium chosen for expression, such as *C. glutamicum* (Wada et al., 1992 Nucleic Acids Res. 20:2111-2118). Such alteration of nucleic acid sequences of the invention can be carried out by standard DNA synthesis techniques.

In another embodiment, the PKSRP expression vector is a yeast expression vector. Examples of vectors for expression in yeast *S. cerevisiae* include pYepSec1 (Baldari, et al., 1987 Embo J. 6:229-234), pMFa (Kurjan and Herskowitz, 1982 Cell 30:933-943), pJRY88 (Schultz et al., 1987 Gene 54:113-123), and pYES2 (Invitrogen Corporation, San Diego, Calif.). Vectors and methods for the construction of vectors appropriate for use in other fungi, such as the filamentous fungi, include those detailed in: van den Hondel, C. A. M. J. J. & Punt, P. J. (1991) "Gene transfer systems and vector development for filamentous fungi, in: Applied Molecular Genetics of Fungi, J. F. Peberdy, et al., eds., p. 1-28, Cambridge University Press: Cambridge.

Alternatively, the PKSRPs of the invention can be expressed in insect cells using baculovirus expression vectors. Baculovirus vectors available for expression of proteins in cultured insect cells (e.g., Sf9 cells) include the pAc series (Smith et al., 1983 Mol. Cell Biol. 3:2156-2165) and the pVL series (Lucklow and Summers, 1989 Virology 170:31-39).

In yet another embodiment, a PKSRP nucleic acid of the invention is expressed in mammalian cells using a mammalian expression vector. Examples of mammalian expression vectors include pCDM8 (Seed, B., 1987 Nature 329:840) and pMT2PC (Kaufman et al., 1987 EMBO J. 6:187-195). When used in mammalian cells, the expression vector's control functions are often provided by viral regulatory elements. For example, commonly used promoters are derived from polyoma, Adenovirus 2, cytomegalovirus and Simian Virus 40. For other suitable expression systems for both prokaryotic and eukaryotic cells see chapters 16 and 17 of Sambrook, J., Fritsch, E. F., and Maniatis, T. *Molecular Cloning: A Laboratory Manual*, 2<sup>nd</sup>, ed., Cold Spring Harbor Laboratory, Cold Spring Harbor Laboratory Press, Cold Spring Harbor, N.Y., 1989.

In another embodiment, the recombinant mammalian expression vector is capable of directing expression of the nucleic acid preferentially in a particular cell type (e.g., tissue-specific regulatory elements are used to express the nucleic acid). Tissue-specific regulatory elements are known in the art. Non-limiting examples of suitable tissue-specific promoters include the albumin promoter (liver-specific; Pinkert et al., 1987 Genes Dev. 1:268-277), lymphoid-specific promoters (Calame and Eaton, 1988 Adv. Immunol. 43:235-275), in particular promoters of T cell receptors (Winoto and Baltimore, 1989 EMBO J. 8:729-733) and immunoglobulins (Banerji et al., 1983 Cell 33:729-740; Queen and Baltimore, 1983 Cell 33:741-748), neuron-specific promoters (e.g., the neurofilament promoter; Byrne and Ruddle, 1989 PNAS 86:5473-5477), pancreas-specific promoters (Edlund et al., 1985 Science 230:912-916), and mammary gland-specific promoters (e.g., milk whey promoter; U.S. Pat. No. 4,873,316 and European Application Publication No. 264,166). Developmentally-regulated promoters are also encompassed, for example, the murine hox promoters (Kessel and Gruss, 1990 Science 249:374-379) and the fetoprotein promoter (Campes and Tilghman, 1989 Genes Dev. 3:537-546).

In another embodiment, the PKSRPs of the invention may be expressed in unicellular plant cells (such as algae) (see Faleiatiore et al., 1999 Marine Biotechnology 1(3):239-251 and references therein) and plant cells from higher plants (e.g., the spermatophytes, such as crop plants). Examples of

plant expression vectors include those detailed in: Becker, D., Kemper, E., Schell, J. and Masterson, R., 1992 New plant binary vectors with selectable markers located proximal to the left border, Plant Mol. Biol. 20: 1195-1197; and Bevan, M. W., 1984 Binary *Agrobacterium* vectors for plant transformation, Nucl. Acid. Res. 12:8711-8721; Vectors for Gene Transfer in Higher Plants; in: Transgenic Plants, Vol. 1, Engineering and Utilization, eds.: Kung and R. Wu, Academic Press, 1993, S. 15-38.

A plant expression cassette preferably contains regulatory sequences capable of driving gene expression in plant cells and operably linked so that each sequence can fulfill its function, for example, termination of transcription by polyadenylation signals. Preferred polyadenylation signals are those originating from *Agrobacterium tumefaciens* t-DNA such as the gene 3 known as octopine synthase of the Ti-plasmid pTiACH5 (Gielen et al., 1984 EMBO J. 3:835) or functional equivalents thereof but also all other terminators functionally active in plants are suitable,

As plant gene expression is very often not limited on transcriptional levels, a plant expression cassette preferably contains other operably linked sequences like translational enhancers such as the overdrive-sequence containing the 5'-untranslated leader sequence from tobacco mosaic virus enhancing the protein per RNA ratio (Gallie et al., 1987 Nucl. Acids Research 15:8693-8711).

Plant gene expression has to be operably linked to an appropriate promoter conferring gene expression in a timely, cell or tissue specific manner. Preferred are promoters driving constitutive expression (Benfey et al., 1989 EMBO J. 8:2195-2202) like those derived from plant viruses like the 35S CAMV (Franck et al., 1980 Cell 21:285-294), the 19S CaMV (see also U.S. Pat. No. 5,352,605 and PCT Application No, WO 8402913) or plant promoters like those from Rubisco small subunit described in U.S. Pat. No. 4,962,028.

Other preferred sequences for use in plant gene expression cassettes are targeting-sequences necessary to direct the gene product in its appropriate cell compartment (for review see Kermod, 1996 Crit. Rev. Plant Sci. 15(4):285-423 and references cited therein) such as the vacuole, the nucleus, all types of plastids like amyloplasts, chloroplasts, chromoplasts, the extracellular space, mitochondria, the endoplasmic reticulum, oil bodies, peroxisomes and other compartments of plant cells.

Plant gene expression can also be facilitated via an inducible promoter (for review see Gatz, 1997 Annu. Rev. Plant Physiol. Plant Mol. Biol. 48:89-108). Chemically inducible promoters are especially suitable if gene expression is wanted to occur in a time specific manner. Examples of such promoters are a salicylic acid inducible promoter (PCT Application No. WO 95/19443), a tetracycline inducible promoter (Gatz et al., 1992 Plant J. 2:397-404) and an ethanol inducible promoter (PCT Application No. WO 93/21334).

Also, suitable promoters responding to biotic or abiotic stress conditions are those such as the pathogen inducible PRP1-gene promoter (Ward et al., 1993 Plant. Mol. Biol. 22:361-366), the heat inducible hsp80-promoter from tomato (U.S. Pat. No. 5,187,267), cold inducible alpha-amylase promoter from potato (PCT Application No. WO 96/12814) or the wound-inducible pinII-promoter (European Patent No. 375091). For other examples of drought, cold, and salt-inducible promoters, such as the RD29A promoter, see Yamaguchi-Shinozaki et al. (1993 Mol. Genet. 236:331-340).

Especially preferred are those promoters that confer gene expression in specific tissues and organs, such as guard cells and the root hair cells. Suitable promoters include the napin-gene promoter from rapeseed (U.S. Pat. No. 5,608,152), the

USP-promoter from *Vicia faba* (Baeumlein et al., 1991 Mol Gen Genet. 225(3):459-67), the oleosin-promoter from *Arabidopsis* (PCT Application No. WO 98/45461), the phaseolin-promoter from *Phaseolus vulgaris* (U.S. Pat. No. 5,504,200), the Bce4-promoter from Brassica (PCT Application No. WO 91/13980) or the legumin B4 promoter (LeB4; Baeumlein et al., 1992 Plant Journal, 2(2)233-9) as well as promoters conferring seed specific expression in monocot plants like maize, barley, wheat, rye, rice, etc. Suitable promoters to note are the lpt2 or lpt1-gene promoter from barley (PCT Application No. WO 95/15389 and PCT Application No. WO 95/23230) or those described in PCT Application No. WO 99/16890 (promoters from the barley hordein-gene, rice glutelin gene, rice oryza gene, rice prolamin gene, wheat gliadin gene, wheat glutelin gene, maize zein gene, oat glutelin gene, Sorghum kasirin-gene and rye secalin gene).

Also especially suited are promoters that confer plastid-specific gene expression since plastids are the compartment where lipid biosynthesis occurs. Suitable promoters are the viral RNA-polymerase promoter described in PCT Application No. WO 95/16783 and PCT Application No. WO 97/06250 and the clpP-promoter from *Arabidopsis* described in PCT Application No. WO 99/46394.

The invention further provides a recombinant expression vector comprising a PKSRP DNA molecule of the invention cloned into the expression vector in an antisense orientation. That is, the DNA molecule is operatively linked to a regulatory sequence in a manner that allows for expression (by transcription of the DNA molecule) of an RNA molecule that is antisense to a PKSRP mRNA. Regulatory sequences operatively linked to a nucleic acid molecule cloned in the antisense orientation can be chosen which direct the continuous expression of the antisense RNA molecule in a variety of cell types. For instance, viral promoters and/or enhancers, or regulatory sequences can be chosen which direct constitutive, tissue specific or cell type specific expression of antisense RNA. The antisense expression vector can be in the form of a recombinant plasmid, phagemid or attenuated virus wherein antisense nucleic acids are produced under the control of a high efficiency regulatory region. The activity of the regulatory region can be determined by the cell type into which the vector is introduced. For a discussion of the regulation of gene expression using antisense genes see Weintraub, H. et al., Antisense RNA as a molecular tool for genetic analysis, Reviews-Trends in Genetics, Vol. 1(1) 1986 and Mol et al., 1990 FEBS Letters 268:427-430.

Another aspect of the invention pertains to host cells into which a recombinant expression vector of the invention has been introduced. The terms "host cell" and "recombinant host cell" are used interchangeably herein. It is understood that such terms refer not only to the particular subject cell but they also apply to the progeny or potential progeny of such a cell. Because certain modifications may occur in succeeding generations due to either mutation or environmental influences, such progeny may not, in fact, be identical to the parent cell, but are still included within the scope of the term as used herein.

A host cell can be any prokaryotic or eukaryotic cell. For example, a PKSRP can be expressed in bacterial cells such as *C. glutamicum*, insect cells, fungal cells or mammalian cells (such as Chinese hamster ovary cells (CHO) or COS cells), algae, ciliates, plant cells, fungi or other microorganisms like *C. glutamicum*. Other suitable host cells are known to those skilled in the art.

Vector DNA can be introduced into prokaryotic or eukaryotic cells via conventional transformation or transfection techniques. As used herein, the terms "transformation",

"transfection", "conjugation" and "transduction" are intended to refer to a variety of art-recognized techniques for introducing foreign nucleic acid (e.g., DNA) into a host cell, including calcium phosphate or calcium chloride co-precipitation, DEAF-dextran-mediated transfection, lipofection, natural competence, chemical-mediated transfer and electroporation. Suitable methods for transforming or transfecting host cells including plant cells can be found in Sambrook, et al. (Molecular Cloning: A Laboratory Manual. 2<sup>nd</sup>, ed., Cold Spring Harbor Laboratory, Cold Spring Harbor Laboratory Press, Cold Spring Harbor, N. Y., 1989) and other laboratory manuals such as Methods in Molecular Biology, 1995, Vol. 44, *Agrobacterium* protocols, ed: Gartland and Davey, Humana Press, Totowa, N.J. As biotic and abiotic stress tolerance is a general trait wished to be inherited into a wide variety of plants like maize, wheat, rye, oat, triticale, rice, barley, soybean, peanut, cotton, rapeseed and canola, manihot, pepper, sunflower and tagetes, solanaceous plants like potato, tobacco, eggplant, and tomato, *Vicia* species, pea, alfalfa, bushy plants (coffee, cacao, tea), *Salix* species, trees (oil palm, coconut), perennial grasses and forage crops, these crop plants are also preferred target plants for a genetic engineering as one further embodiment of the present invention,

In particular, the invention provides a method of producing a transgenic plant with a PKSRP coding nucleic acid, wherein expression of the nucleic acid(s) in the plant results in increased tolerance to environmental stress as compared to a wild type variety of the plant comprising: (a) transforming a plant cell with an expression vector comprising a PKSRP nucleic acid, and (b) generating from the plant cell a transgenic plant with a increased tolerance to environmental stress as compared to a wild type variety of the plant. The invention also provides a method of increasing expression of a gene of interest within a host cell as compared to a wild type variety of the host cell, wherein the gene of interest is transcribed in response to a PKSRP, comprising: (a) transforming the host cell with an expression vector comprising a PKSRP coding nucleic acid, and (b) expressing the PKSRP within the host cell, thereby increasing the expression of the gene transcribed in response to the PKSRP, as compared to a wild type variety of the host cell.

For such plant transformation, binary vectors such as pBinAR can be used (Höfgen and Willmitzer, 1990 Plant Science 66:221-230). Construction of the binary vectors can be performed by ligation of the cDNA in sense or antisense orientation into the T-DNA. 5-prime to the cDNA a plant promoter activates transcription of the cDNA. A polyadenylation sequence is located 3-prime to the cDNA. Tissue-specific expression can be achieved by using a tissue specific promoter. For example, seed-specific expression can be achieved by cloning the napin or LeB4 or USP promoter 5-prime to the cDNA. Also, any other seed specific promoter element can be used. For constitutive expression within the whole plant, the CaMV 35S promoter can be used. The expressed protein can be targeted to a cellular compartment using a signal peptide, for example for plastids, mitochondria or endoplasmic reticulum (Kermode, 1996 Crit. Rev. Plant Sci. 4(15):285-423). The signal peptide is cloned 5-prime in frame to the cDNA to archive subcellular localization of the fusion protein. Additionally, promoters that are responsive to abiotic stresses can be used with, such as the *Arabidopsis* promoter RD29A, the nucleic acid sequences disclosed herein. One skilled in the art will recognize that the promoter used should be operatively linked to the nucleic acid such that the promoter causes transcription of the nucleic acid which results in the synthesis of an mRNA which encodes a polypeptide. Alternatively, the



RNA can be an antisense RNA for use in affecting subsequent expression of the same or another gene or genes.

Alternate methods of transfection include the direct transfer of DNA into developing flowers via electroporation or *Agrobacterium* mediated gene transfer. *Agrobacterium* mediated plant transformation can be performed using for example the GV3101(pMP90) (Koncz and Schell, 1986 Mol. Gen. Genet. 204:383-396) or LBA4404 (Clontech) *Agrobacterium tumefaciens* strain. Transformation can be performed by standard transformation and regeneration techniques (Deblaere et al., 1994 Nucl. Acids. Res. 13:4777-4788; Gelvin, Stanton B. and Schilperoort, Robert A, Plant Molecular Biology Manual, 2<sup>nd</sup> Ed.—Dordrecht: Kluwer Academic Publ., 1995.—in Sect., Ringbuc Zentrale Signatur: BT11-P ISBN 0-7923-2731-4; Glick, Bernard R.; Thompson, John E., Methods in Plant Molecular Biology and Biotechnology, Boca Raton: CRC Press, 1991 - 360 S., ISBN 0-8493-5164-2). For example, rapeseed can be transformed via cotyledon or hypocotyl transformation (Moloney et al., 1989 Plant cell Report 8:238-242; De Block et al., 1989 Plant Physiol. 91:694-701). Use of antibiotics for *Agrobacterium* and plant selection depends on the binary vector and the *Agrobacterium* strain used for transformation. Rapeseed selection is normally performed using kanamycin as selectable plant marker. *Agrobacterium* mediated gene transfer to flax can be performed using, for example, a technique described by Mlynarova et al., 1994 Plant Cell Report 13:282-285. Additionally, transformation of soybean can be performed using for example a technique described in European Patent No. 0424 047, U.S. Pat. No. 5,322,783, European Patent No. 0397 687, U.S. Pat. Nos. 5,376,543 or 5,169,770. Transformation of maize can be achieved by particle bombardment, polyethylene glycol mediated DNA uptake or via the silicon carbide fiber technique. (See, for example, Freeling and Walbot "The maize handbook" Springer Verlag: New York (1993) ISBN 3-540-97826-7). A specific example of maize transformation is found in U.S. Pat. No. 5,990,387 and a specific example of wheat transformation can be found in PCT Application No. WO 93/07256.

For stable transfection of mammalian cells, it is known that, depending upon the expression vector and transfection technique used, only a small fraction of cells may integrate the foreign DNA into their genome. In order to identify and select these integrants, a gene that encodes a selectable marker (e.g., resistance to antibiotics) is generally introduced into the host cells along with the gene of interest. Preferred selectable markers include those which confer resistance to drugs, such as G418, hygromycin and methotrexate or in plants that confer resistance towards a herbicide such as glyphosate or glufosinate. Nucleic acid molecules encoding a selectable marker can be introduced into a host cell on the same vector as that encoding a PKSRRP or can be introduced on a separate vector. Cells stably transfected with the introduced nucleic acid molecule can be identified by, for example, drug selection (e.g., cells that have incorporated the selectable marker gene will survive, while the other cells die).

To create a homologous recombinant microorganism, a vector is prepared which contains at least a portion of a PKSRRP gene into which a deletion, addition or substitution has been introduced to thereby alter, e.g., functionally disrupt, the PKSRRP gene. Preferably, the PKSRRP gene is a *Physcomitrella patens* PKSRRP gene, but it can be a homolog from a related plant or even from a mammalian, yeast, or insect source. In a preferred embodiment, the vector is designed such that, upon homologous recombination, the endogenous PKSRRP gene is functionally disrupted (i.e., no longer encodes a functional protein; also referred to as a knock-out vector).

Alternatively, the vector can be designed such that, upon homologous recombination, the endogenous PKSRRP gene is mutated or otherwise altered but still encodes a functional protein (e.g., the upstream regulatory region can be altered to thereby alter the expression of the endogenous PKSRRP). To create a point mutation via homologous recombination, DNA-RNA hybrids can be used in a technique known as chimeraplasty (Cole-Strauss et al., 1999 Nucleic Acids Research 27(5):1323-1330 and Kmiec, 1999 Gene therapy American Scientist. 87(3):240-247). Homologous recombination procedures in *Physcomitrella patens* are also well known in the art and are contemplated for use herein.

Whereas in the homologous recombination vector, the altered portion of the PKSRRP gene is flanked at its 5' and 3' ends by an additional nucleic acid molecule of the PKSRRP gene to allow for homologous recombination to occur between the exogenous PKSRRP gene carried by the vector and an endogenous PKSRRP gene, in a microorganism or plant. The additional flanking PKSRRP nucleic acid molecule is of sufficient length for successful homologous recombination with the endogenous gene. Typically, several hundreds of base pairs up to kilobases of flanking DNA (both at the 5' and 3' ends) are included in the vector (see e.g., Thomas, K. R., and Capecchi, M. R., 1987 Cell 51:503 for a description of homologous recombination vectors or Strepp et al., 1998 PNAS, 95 (8):4368-4373 for cDNA based recombination in *Physcomitrella patens*). The vector is introduced into a microorganism or plant cell (e.g., via polyethylene glycol mediated DNA), and cells in which the introduced PKSRRP gene has homologously recombined with the endogenous PKSRRP gene are selected using art-known techniques.

In another embodiment, recombinant microorganisms can be produced that contain selected systems which allow for regulated expression of the introduced gene. For example, inclusion of a PKSRRP gene on a vector placing it under control of the lac operon permits expression of the PKSRRP gene only in the presence of IPTG. Such regulatory systems are well known in the art.

A host cell of the invention, such as a prokaryotic or eukaryotic host cell in culture, can be used to produce (i.e., express) a PKSRRP. Accordingly, the invention further provides methods for producing PKSRRPs using the host cells of the invention. In one embodiment, the method comprises culturing the host cell of invention (into which a recombinant expression vector encoding a PKSRRP has been introduced, or into which genome has been introduced a gene encoding a wild-type or altered PKSRRP) in a suitable medium until PKSRRP is produced. In another embodiment, the method further comprises isolating PKSRRPs from the medium or the host cell.

Another aspect of the invention pertains to isolated PKSRRPs, and biologically active portions thereof. An "isolated" or "purified" protein or biologically active portion thereof is free of some of the cellular material when produced by recombinant DNA techniques, or chemical precursors or other chemicals when chemically synthesized. The language "substantially free of cellular material" includes preparations of PKSRRP in which the protein is separated from some of the cellular components of the cells in which it is naturally or recombinantly produced. In one embodiment, the language "substantially free of cellular material" includes preparations of a PKSRRP having less than about 30% (by dry weight) of non-PKSRRP material (also referred to herein as a "contaminating protein"), more preferably less than about 20% of non-PKSRRP material, still more preferably less than about 10% of non-PKSRRP material, and most preferably less than about 5% non-PKSRRP material.

When the PKSRP or biologically active portion thereof is recombinantly produced, it is also preferably substantially free of culture medium, i.e., culture medium represents less than about 20%, more preferably less than about 10%, and most preferably less than about 5% of the volume of the protein preparation. The language "substantially free of chemical precursors or other chemicals" includes preparations of PKSRP in which the protein is separated from chemical precursors or other chemicals that are involved in the synthesis of the protein. In one embodiment, the language "substantially free of chemical precursors or other chemicals" includes preparations of a PKSRP having less than about 30% (by dry weight) of chemical precursors or non-PKSRP chemicals, more preferably less than about 20% chemical precursors or non-PKSRP chemicals, still more preferably less than about 10% chemical precursors or non-PKSRP chemicals, and most preferably less than about 5% chemical precursors or non-PKSRP chemicals. In preferred embodiments, isolated proteins, or biologically active portions thereof, lack contaminating proteins from the same organism from which the PKSRP is derived. Typically, such proteins are produced by recombinant expression of, for example, a *Physcomitrella patens* PKSRP in plants other than *Physcomitrella patens* or microorganisms such as *C. glutamicum*, ciliates, algae or fungi.

The nucleic acid molecules, proteins, protein homologs, fusion proteins, primers, vectors, and host cells described herein can be used in one or more of the following methods: identification of *Physcomitrella patens* and related organisms; mapping of genomes of organisms related to *Physcomitrella patens*; identification and localization of *Physcomitrella patens* sequences of interest; evolutionary studies; determination of PKSRP regions required for function; modulation of a PKSRP activity; modulation of the metabolism of one or more cell functions; modulation of the transmembrane transport of one or more compounds; and modulation of stress resistance.

The moss *Physcomitrella patens* represents one member of the mosses. It is related to other mosses such as *Ceratodon purpureus* which is capable of growth in the absence of light. Mosses like *Ceratodon* and *Physcomitrella* share a high degree of homology on the DNA sequence and polypeptide level allowing the use of heterologous screening of DNA molecules with probes evolving from other mosses or organisms, thus enabling the derivation of a consensus sequence suitable for heterologous screening or functional annotation and prediction of gene functions in third species. The ability to identify such functions can therefore have significant relevance, e.g., prediction of substrate specificity of enzymes. Further, these nucleic acid molecules may serve as reference points for the mapping of moss genomes, or of genomes of related organisms.

The PKSRP nucleic acid molecules of the invention have a variety of uses. Most importantly, the nucleic acid and amino acid sequences of the present invention can be used to transform plants, thereby inducing tolerance to stresses such as drought, high salinity and cold. The present invention therefore provides a transgenic plant transformed by a PKSRP nucleic acid (coding or antisense), wherein expression of the nucleic acid sequence in the plant results in increased tolerance to environmental stress as compared to a wild type variety of the plant. The transgenic plant can be a monocot or a dicot. The invention further provides that the transgenic plant can be selected from maize, wheat, lye, oat, triticale, rice, barley, soybean, peanut, cotton, rapeseed, canola, manihot, pepper, sunflower, tagetes, solanaceous plants, potato, tobacco, eggplant, tomato, *Vicia* species, pea, alfalfa, coffee,

cacao, tea, *Salix* species, oil palm, coconut, perennial grass and forage crops, for example.

In particular, the present invention describes using the expression of PK-6, PK-7, PK-8, PK-9, CK-1, CK-2, CK-3, MPK-2, MPK-3, MPK-4, MPK-5, CPK-1 and CPK-2 of *Physcomitrella patens* to engineer drought-tolerant, salt-tolerant and/or cold-tolerant plants. This strategy has herein been demonstrated for *Arabidopsis thaliana*, Rapeseed/Canola, soybeans, corn and wheat but its application is not restricted to these plants. Accordingly, the invention provides a transgenic plant containing a PKSRP selected from PK-6 (SEQ ID NO:27), PK-7 (SEQ ID NO:28), PK-8 (SEQ ID NO:29), PK-9 (SEQ ID NO:30), CK-1 (SEQ ID NO:31), CK-2 (SEQ ID NO:32), CK-3 (SEQ ID NO:33), MPK-2 (SEQ ID NO:34), MPK-3 (SEQ ID NO:35), MPK-4 (SEQ ID NO:36), MPK-5 (SEQ ID NO:37), CPK-1 (SEQ ID NO:38) and CPK-2 (SEQ ID NO:39), wherein the environmental stress is drought, increased salt or decreased or increased temperature. In preferred embodiments, the environmental stress is drought or decreased temperature.

The present invention also provides methods of modifying stress tolerance of a plant comprising, modifying the expression of a PKSRP in the plant. The invention provides that this method can be performed such that the stress tolerance is either increased or decreased. In particular, the present invention provides methods of producing a transgenic plant having an increased tolerance to environmental stress as compared to a wild type variety of the plant comprising increasing expression of a PKSRP in a plant.

The methods of increasing expression of PKSRPs can be used wherein the plant is either transgenic or not transgenic. In cases when the plant is transgenic, the plant can be transformed with a vector containing any of the above described PKSRP coding nucleic acids, or the plant can be transformed with a promoter that directs expression of native PKSRP in the plant, for example. The invention provides that such a promoter can be tissue specific. Furthermore, such a promoter can be developmentally regulated. Alternatively, non-transgenic plants can have native PKSRP expression modified by inducing a native promoter.

The expression of PK-6 (SEQ ID NO:14), PK-7 (SEQ ID NO:15), PK-8 (SEQ ID NO:16), PK-9 (SEQ ID NO:17), CK-1 (SEQ ID NO:18), CK-2 (SEQ ID NO:19), CK-3 (SEQ ID NO:20), MPK-2 (SEQ ID NO:21), MPK-3 (SEQ ID NO:22), MPK-4 (SEQ ID NO:23), MPK-5 (SEQ ID NO:24), CPK-1 (SEQ ID NO:25) and CPK-2 (SEQ ID NO:26) in target plants can be accomplished by, but is not limited to, one of the following examples: (a) constitutive promoter, (b) stress-inducible promoter, (c) chemical-induced promoter, and (d) engineered promoter over-expression with for example zinc-finger derived transcription factors (Greisman and Pabo, 1997 Science 275:657). The later case involves identification of the PK-6 (SEQ ID NO:27), PK-7 (SEQ ID NO:28), PK-8 (SEQ ID NO:29), PK-9 (SEQ ID NO:30), CK-1 (SEQ ID NO:31), CK-2 (SEQ ID NO:32), CK-3 (SEQ ID NO:33), MPK-2 (SEQ ID NO:34), MPK-3 (SEQ ID NO:35), MPK-4 (SEQ ID NO:36), MPK-5 (SEQ ID NO:37), CPK-1 (SEQ ID NO:38) or CPK-2 (SEQ ID NO:39) homologs in the target plant as well as from its promoter. Zinc-finger-containing recombinant transcription factors are engineered to specifically interact with the PK-6 (SEQ ID NO:27), PK-7 (SEQ ID NO:28), PK-8 (SEQ ID NO:29), PK-9 (SEQ ID NO:30), CK-1 (SEQ ID NO:31), CK-2 (SEQ ID NO:32), CK-3 (SEQ ID NO:33), MPK-2 (SEQ ID NO:34), MPK-3 (SEQ ID NO:35), MPK-4 (SEQ ID NO:36),

MPK-5 (SEQ ID NO:37), CPK-1 (SEQ ID NO:38) or CPK-2 (SEQ ID NO:39) homolog and transcription of the corresponding gene is activated.

In addition to introducing the PKSRP nucleic acid sequences into transgenic plants, these sequences can also be used to identify an organism as being *Physcomitrella patens* or a close relative thereof. Also, they may be used to identify the presence of *Physcomitrella patens* or a relative thereof in a mixed population of microorganisms. The invention provides the nucleic acid sequences of a number of *Physcomitrella patens* genes; by probing the extracted genomic DNA of a culture of a unique or mixed population of microorganisms under stringent conditions with a probe spanning a region of a *Physcomitrella patens* gene which is unique to this organism, one can ascertain whether this organism is present.

Further, the nucleic acid and protein molecules of the invention may serve as markers for specific regions of the genome. This has utility not only in the mapping of the genome, but also in functional studies of *Physcomitrella patens* proteins. For example, to identify the region of the genome to which a particular *Physcomitrella patens* DNA-binding protein binds, the *Physcomitrella patens* genome could be digested, and the fragments incubated with the DNA-binding protein. Those fragments that bind the protein may be additionally probed with the nucleic acid molecules of the invention, preferably with readily detectable labels. Binding of such a nucleic acid molecule to the genome fragment enables the localization of the fragment to the genome map of *Physcomitrella patens*, and, when performed multiple times with different enzymes, facilitates a rapid determination of the nucleic acid sequence to which the protein binds. Further, the nucleic acid molecules of the invention may be sufficiently homologous to the sequences of related species such that these nucleic acid molecules may serve as markers for the construction of a genomic map in related mosses,

The PKSRP nucleic acid molecules of the invention are also useful for evolutionary and protein structural studies. The metabolic and transport processes in which the molecules of the invention participate are utilized by a wide variety of prokaryotic and eukaryotic cells; by comparing the sequences of the nucleic acid molecules of the present invention to those encoding similar enzymes from other organisms, the evolutionary relatedness of the organisms can be assessed. Similarly, such a comparison permits an assessment of which regions of the sequence are conserved and which are not, which may aid in determining those regions of the protein that are essential for the functioning of the enzyme. This type of determination is of value for protein engineering studies and may give an indication of what the protein can tolerate in terms of mutagenesis without losing function.

Manipulation of the PKSRP nucleic acid molecules of the invention may result in the production of PKSRPs having functional differences from the wild-type PKSRPs. These proteins may be improved in efficiency or activity, may be present in greater numbers in the cell than is usual, or may be decreased in efficiency or activity.

There are a number of mechanisms by which the alteration of a PKSRP of the invention may directly affect stress response and/or stress tolerance. In the case of plants expressing PKSRPs, increased transport can lead to improved salt and/or solute partitioning within the plant tissue and organs. By either increasing the number or the activity of transporter molecules which export ionic molecules from the cell, it may be possible to affect the salt tolerance of the cell.

The effect of the genetic modification in plants, *C. glutamicum*, fungi, algae, or ciliates on stress tolerance can be assessed by growing the modified microorganism or plant

under less than suitable conditions and then analyzing the growth characteristics and/or metabolism of the plant. Such analysis techniques are well known to one skilled in the art, and include dry weight, wet weight, protein synthesis, carbohydrate synthesis, lipid synthesis, evapotranspiration rates, general plant and/or crop yield, flowering, reproduction, seed setting, root growth, respiration rates, photosynthesis rates, etc. (Applications of HPLC in Biochemistry in Laboratory Techniques in Biochemistry and Molecular Biology, vol. 17; Rehm et al., 1993 Biotechnology, vol. 3, Chapter III: Product recovery and purification, page 469-714, VCH: Weinheim; Belter, P. A. et al., 1988 Bioseparations: downstream processing for biotechnology, John Wiley and Sons; Kennedy, J. F. and Cabral, J. M. S., 1992 Recovery processes for biological materials, John Wiley and Sons; Shaeiwitz, J. A. and Hemy, J. D., 1988 Biochemical separations, in: Ulmann's Encyclopedia of Industrial Chemistry, vol. B3, Chapter 11, page 1-27, VCH: Weinheim; and Dechow, F. J. (1989) Separation and purification techniques in biotechnology, Noyes Publications).

For example, yeast expression vectors comprising the nucleic acids disclosed herein, or fragments thereof, can be constructed and transformed into *Saccharomyces cerevisiae* using standard protocols. The resulting transgenic cells can then be assayed for fail or alteration of their tolerance to drought, salt, and temperature stress. Similarly, plant expression vectors comprising the nucleic acids disclosed herein, or fragments thereof, can be constructed and transformed into an appropriate plant cell such as *Arabidopsis*, soy, rape, maize, wheat, *Medicago truncatula*, etc., using standard protocols. The resulting transgenic cells and/or plants derived therefrom can then be assayed for fail or alteration of their tolerance to drought, salt, and temperature stress.

The engineering of one or more PKSRP genes of the invention may also result in PKSRPs having altered activities which indirectly impact the stress response and/or stress tolerance of algae, plants, ciliates or fungi or other microorganisms like *C. glutamicum*. For example, the normal biochemical processes of metabolism result in the production of a variety of products (e.g., hydrogen peroxide and other reactive oxygen species) which may actively interfere with these same metabolic processes (for example, peroxyxynitrite is known to nitrate tyrosine side chains, thereby inactivating some enzymes having tyrosine in the active site (Groves, J. T., 1999 Curr. Opin. Chem. Biol. 3(2):226-235). While these products are typically excreted, cells can be genetically altered to transport more products than is typical for a wild-type cell. By optimizing the activity of one or more PKSRPs of the invention which are involved in the export of specific molecules, such as salt molecules, it may be possible to improve the stress tolerance of the cell.

Additionally, the sequences disclosed herein, or fragments thereof, can be used to generate knockout mutations in the genomes of various organisms, such as bacteria, mammalian cells, yeast cells, and plant cells (Girke, T., 1998 The Plant Journal 15:39-48). The resultant knockout cells can then be evaluated for their ability or capacity to tolerate various stress conditions, their response to various stress conditions, and the effect on the phenotype and/or genotype of the mutation. For other methods of gene inactivation see U.S. Pat. No. 6,004, 804 "Non-Chimeric Mutational Vectors" and Puttaraju et al., 1999 Spliceosome-mediated RNA trans-splicing as a tool for gene therapy Nature Biotechnology 17:246-252.

The aforementioned mutagenesis strategies for PKSRPs resulting in increased stress resistance are not meant to be limiting; variations on these strategies will be readily apparent to one skilled in the art. Using such strategies, and incor-

porating the mechanisms disclosed herein, the nucleic acid and protein molecules of the invention may be utilized to generate algae, ciliates, plants, fungi or other microorganisms like *C. glutamicum* expressing mutated PKSRP nucleic acid and protein molecules such that the stress tolerance is improved.

The present invention also provides antibodies that specifically bind to a PKSRP, or a portion thereof, as encoded by a nucleic acid described herein. Antibodies can be made by many well-known methods (See, e.g. Harlow and Lane, "Antibodies; A Laboratory Manual" Cold Spring Harbor Laboratory, Cold Spring Harbor, N.Y., (1988)). Briefly, purified antigen can be injected into an animal in an amount and in intervals sufficient to elicit an immune response. Antibodies can either be purified directly, or spleen cells can be obtained from the animal. The cells can then fused with an immortal cell line and screened for antibody secretion. The antibodies can be used to screen nucleic acid clone libraries for cells secreting the antigen. Those positive clones can then be sequenced. (See, for example, Kelly et al., 1992 Bio/Technology 10:163-167; Bebbington et al., 1992 Bio/Technology 10:169-175).

The phrases "selectively binds" and "specifically binds" with the polypeptide refer to a binding reaction that is determinative of the presence of the protein in a heterogeneous population of proteins and other biologics. Thus, under designated immunoassay conditions, the specified antibodies bound to a particular protein do not bind in a significant amount to other proteins present in the sample. Selective binding of an antibody under such conditions may require an antibody that is selected for its specificity for a particular protein. A variety of immunoassay formats may be used to select antibodies that selectively bind with a particular protein. For example, solid-phase ELISA immunoassays are routinely used to select antibodies selectively immunoreactive with a protein. See Harlow and Lane "Antibodies, A Laboratory Manual" Cold Spring Harbor Publications, New York, (1988), for a description of immunoassay formats and conditions that could be used to determine selective binding.

In some instances, it is desirable to prepare monoclonal antibodies from various hosts. A description of techniques for preparing such monoclonal antibodies may be found in Stites et al., editors, "Basic and Clinical Immunology," (Lange Medical Publications, Los Altos, Calif., Fourth Edition) and references cited therein, and in Harlow and Lane ("Antibodies, A Laboratory Manual" Cold Spring Harbor Publications, New York, 1988).

Throughout this application, various publications are referenced. The disclosures of all of these publications and those references cited within those publications in their entireties are hereby incorporated by reference into this application in order to more fully describe the state of the art to which this invention pertains.

It should also be understood that the foregoing relates to preferred embodiments of the present invention and that numerous changes may be made therein without departing from the scope of the invention. The invention is further illustrated by the following examples, which are not to be construed in any way as imposing limitations upon the scope thereof. On the contrary, it is to be clearly understood that resort may be had to various other embodiments, modifications, and equivalents thereof; which, after reading the description herein, may suggest themselves to those skilled in the art without departing from the spirit of the present invention and/or the scope of the appended claims.

## Example 1

5 Growth of *Physcomitrella Patens* Cultures

For this study, plants of the species *Physcomitrella patens* (Hedw.) B.S.G. from the collection of the genetic studies section of the University of Hamburg were used. They originate from the strain 16/14 collected by H. L. K. Whitehouse in Gransden Wood, Huntingdonshire (England), which was subcultured from a spore by Engel (1968, Am. J. Bot. 55, 438-446). Proliferation of the plants was carried out by means of spores and by means of regeneration of the gametophytes. The protonema developed from the haploid spore as a chloroplast-rich chloronema and chloroplast-low caulonema, on which buds formed after approximately 12 days. These grew to give gametophores bearing antheridia and archegonia. After fertilization, the diploid sporophyte with a short seta and the spore capsule resulted, in which the meiospores matured.

Culturing was carried out in a climatic chamber at an air temperature of 25° C. and light intensity of 55 micromol s<sup>-1</sup> m<sup>-2</sup> (white light; Philips TL 65W/25 fluorescent tube) and a light/dark change of 16/8 hours. The moss was either modified in liquid culture using Knop medium according to Reski and Abel (1985, Planta 165:354-358) or cultured on Knop solid medium using 1% oxoid agar (Unipath, Basingstoke, England). The protonemas used for RNA and DNA isolation were cultured in aerated liquid cultures. The protonemas were comminuted every 9 days and transferred to fresh culture medium.

## Example 2

## 35 Total DNA Isolation from Plants

The details for the isolation of total DNA relate to the working up of one gram fresh weight of plant material. The materials used include the following buffers: CTAB buffer: 2% (w/v) N-cethyl-N,N,N-trimethylammonium bromide (CTAB); 100 mM Tris HCl pH 8.0; 1.4 M NaCl; 20 mM EDTA; N-Laurylsarcosine buffer: 10% (w/v) N-laurylsarcosine; 100 mM Tris HCl pH 8.0; 20 mM EDTA.

The plant material was triturated under liquid nitrogen in a mortar to give a fine powder and transferred to 2 ml Eppendorf vessels. The frozen plant material was then covered with a layer of 1 ml of decomposition buffer (1 ml CTAB buffer, 100 µl of N-laurylsarcosine buffer, 20 µl of β-mercaptoethanol and 10 µl of proteinase K solution, 10 mg/ml) and incubated at 60° C. for one hour with continuous shaking. The homogenate obtained was distributed into two Eppendorf vessels (2 ml) and extracted twice by shaking with the same volume of chloroform/isoamyl alcohol (24:1). For phase separation, centrifugation was carried out at 8000×g and room temperature for 15 minutes in each case. The DNA was then precipitated at -70° C. for 30 minutes using ice-cold isopropanol. The precipitated DNA was sedimented at 4° C. and 10,000 g for 30 minutes and resuspended in 180 µl of TE buffer (Sambrook et al., 1989, Cold Spring Harbor Laboratory Press: ISBN 0-87969-309-6). For further purification, the DNA was treated with NaCl (1.2 M final concentration) and precipitated again at -70° C. for 30 minutes using twice the volume of absolute ethanol. After a washing step with 70% ethanol, the DNA was dried and subsequently taken up in 50 µl of H<sub>2</sub>O+RNase (50 mg/ml final concentration). The DNA was dissolved overnight at 4° C. and the RNase digestion was subsequently carried out at 37° C. for 1 hour. Storage of the DNA took place at 4° C.

Isolation of Total RNA and poly-(A)+ RNA and cDNA Library Construction from *Physcomitrella Patens*

For the investigation of transcripts, both total RNA and poly-(A)+ RNA were isolated. The total RNA was obtained from wild-type 9 day old protonemata following the GTC-method (Reski et al. 1994, Mol. Gen. Genet., 244:352-359). The Poly(A)+ RNA was isolated using Dyna Beads<sup>R</sup> (Dyna, Oslo, Norway) following the instructions of the manufacturers protocol. After determination of the concentration of the RNA or of the poly(A)+ RNA, the RNA was precipitated by addition of 1/10 volumes of 3 M sodium acetate pH 4.6 and 2 volumes of ethanol and stored at -70° C.

For cDNA library construction, first strand synthesis was achieved using Murine Leukemia Virus reverse transcriptase (Roche, Mannheim, Germany) and oligo-d(T)-primers, second strand synthesis by incubation with DNA polymerase I, Klenow enzyme and RNaseH digestion at 12° C. (2 hours), 16° C. (1 hour) and 22° C. (1 hour). The reaction was stopped by incubation at 65° C. (10 minutes) and subsequently transferred to ice. Double stranded DNA molecules were blunted by T4-DNA-polymerase (Roche, Mannheim) at 37° C. (30 minutes). Nucleotides were removed by phenol/chloroform extraction and Sephadex G50 spin columns. EcoRI adapters (Pharmacia, Freiburg, Germany) were ligated to the cDNA ends by T4-DNA-ligase (Roche, 12° C., overnight) and phosphorylated by incubation with polynucleotide kinase (Roche, 37° C., 30 minutes). This mixture was subjected to separation on a low melting agarose gel. DNA molecules larger than 300 base pairs were eluted from the gel, phenol extracted, concentrated on Elutip-D-columns (Schleicher and Schuell, Dassel, Germany) and were ligated to vector arms and packed into lambda ZAPII phages or lambda ZAP-Express phages using the Gigapack Gold Kit (Stratagene, Amsterdam, Netherlands) using material and following the instructions of the manufacturer.

## Example 4

Sequencing and Function Annotation of *Physcomitrella Patens* ESTs

cDNA libraries as described in Example 3 were used for DNA sequencing according to standard methods, and in particular, by the chain termination method using the ABI PRISM Big Dye Terminator Cycle Sequencing Ready Reaction Kit (Perkin-Elmer, Weiterstadt, Germany). Random Sequencing was carried out subsequent to preparative plasmid recovery from cDNA libraries via in vivo mass excision, retransformation, and subsequent plating of DH10B on agar plates (material and protocol details from Stratagene, Amsterdam, Netherlands). Plasmid DNA was prepared from overnight grown *E. coli* cultures grown in Luria-Broth medium containing ampicillin (see Sambrook et al. 1989 Cold Spring Harbor Laboratory Press: ISBN 0-87969-309-6) on a Qiagen DNA preparation robot (Qiagen, Hilden) according to the manufacturer's protocols. Sequencing primers with the following nucleotide sequences were used:

5' - CAGGAACAGCTATGACC - 3'      SEQ ID NO: 40  
 5' - CTAAGGGAACAAAAGCTG - 3'      SEQ ID NO: 41  
 5' - TGTAACACGACGCCAGT - 3'      SEQ ID NO: 42

Sequences were processed and annotated using the software package EST-MAX commercially provided by BioMax (Munich, Germany). The program incorporates practically all bioinformatics methods important for functional and structural characterization of protein sequences. For reference the website at pedant.nzips.biochem.mpg.de. The most important algorithms incorporated in EST-MAX are: FASTA: Very sensitive sequence database searches with estimates of statistical significance; Pearson W. R. (1990) Rapid and sensitive sequence comparison with FASTP and FASTA. Methods Enzymol. 183:63-98; BLAST: Very sensitive sequence database searches with estimates of statistical significance. Altschul S. F., Gish W., Miller W., Myers E. W., and Lipman D. J. Basic local alignment search tool. Journal of Molecular Biology 215:403-10; PREDATOR: High-accuracy secondary structure prediction from single and multiple sequences. Frishman, D. and Argos, P. (1997) 75% accuracy in protein secondary structure prediction. Proteins, 27:329-335; CLUSTALW: Multiple sequence alignment. Thompson, J. D., Higgins, D. G. and Gibson, T. J. (1994) CLUSTAL W: improving the sensitivity of progressive multiple sequence alignment through sequence weighting, positions-specific gap penalties and weight matrix choice. Nucleic Acids Research, 22:4673-4680; TMAP: Transmembrane region prediction from multiply aligned sequences. Persson, B. and Argos, P. (1994) Prediction of transmembrane segments in proteins utilizing multiple sequence alignments. J. Mol. Biol. 237:182-192; ALOM2: Transmembrane region prediction from single sequences. Klein, P., Kanehisa, M., and DeLisi, C. Prediction of protein function from sequence properties: A discriminate analysis of a database, Biochim. Biophys. Acta 787:221-226 (1984). Version 2 by Dr. K. Nakai; PROSE-ARCH: Detection of PROSITE protein sequence patterns. Kolakowski L. F. Jr., Leunissen J. A. M., Smith J. E. (1992) ProSearch: fast searching of protein sequences with regular expression patterns related to protein structure and function. Biotechniques 13, 919-921; BLIMPS: Similarity searches against a database of ungapped blocks, J. C. Wallace and Henikoff S., (1992); PATMAT: A searching and extraction program for sequence, pattern and block queries and databases, CABIOS 8:249-254. Written by Bill Alford.

## Example 5

Identification of *Physcomitrella Patens* ORFs Corresponding to PK-6, PK-7, PK-8, PK-9, CK-1, CK-2, CK-3, MPK-2, MPK-4, MPK-5, CPK-1 and CPK-2

The *Physcomitrella patens* partial cDNAs (ESTs) shown in Table 1 below were identified in the *Physcomitrella patens* EST sequencing program using the program EST-MAX through BLAST analysis. The Sequence Identification Numbers corresponding to these ESTs are as follows: PK-6 (SEQ ID NO:1), PK-7 (SEQ ID NO:2), PK-8 (SEQ ID NO:3), PK-9 (SEQ ID NO:4), CK-1 (SEQ ID NO:5), CK-2 (SEQ ID NO:6), CK-3 (SEQ ID NO:7), MPK-2 (SEQ ID NO:8), MPK-3 (SEQ ID NO:9), MPK-4 (SEQ ID NO:10), MPK-5 (SEQ ID NO:11), CPK-1 (SEQ ID NO:12) and CPK-2 (SEQ ID NO:13).

TABLE 1

Name	Functional categories	Function	Sequence code	ORF position
PpPK-6	Protein Kinase	serine/threonine protein kinase like protein	c_pp004044242r	1-474
PpPK-7	Protein Kinase	cdc2-like protein kinase cdc2MsF	s_pp001031042f	1-267
PpPK-8	Protein Kinase	protein kinase homolog F13C5.120	c_pp004044100r	1-581
PpPK-9	Protein Kinase	protein kinase; similar to human PKX1	c_pp004071077r	709-137
PpCK-1	Protein Kinase	receptor protein kinase	c_pp001062017r	1160-1
PpCK-2	Protein Kinase	kasein kinase	c_pp004038371r	1909-1421
PpCK-3	Protein Kinase	casein kinase II catalytic subunit	c_pp004076164r	2-877
PpMPK-2	Protein Kinase	mitogen-activated protein kinase 6	c_pp004041329r	952-293
PpMPK-3	Protein Kinase	big MAP kinase 1c	c_pp004061263r	221-550
PpMPK-4	Protein Kinase	protein kinase MEK1 (EC 2.7.1.-)	c_pp001064077r	1153-596
PpMPK-5	Protein Kinase	protein kinase MEK1	c_pp004064129r	114-233
PpCPK-1	Protein Kinase	protein kinase	c_pp004014376r	1084-173
PpCPK-2	Protein Kinase	calcium-dependent protein kinase	c_pp004038141r	422-1213
PpPK-6	Protein Kinase	cdc2-like protein kinase cdc2MsF	s_pp001031042f	1-267

TABLE 2

Degree of Amino Acid Identity and Similarity of PpPK-6 and Other Homologous Proteins GCG Gap program was used: gap penalty: 10; gap extension penalty: 0.1; score matrix: blosum62)					
Swiss-Prot #					
	O81106	Q9LUL4	Q9ZQZ2	Q9MAS2	Q9LK66
Protein name	LEUCINE-RICH REPEAT TRANS-MEMBRANE PROTEIN KINASE 2	SERINE/THREONINE PROTEIN KINASE LIKE PROTEIN	PUTATIVE LRR LINKED PROTEIN KINASE	PUTATIVE LRR RECEPTOR PROTEIN KINASE	PROTEIN KINASE-LIKE PROTEIN
Species	<i>Zea mays</i> (Maize)	<i>Arabidopsis thaliana</i> (Mouse-ear cress)	<i>A. thaliana</i>	<i>A. thaliana</i>	<i>A. thaliana</i>
Identity %	42%	42%	38%	37%	37%
Similarity %	54%	52%	50%	49%	48%

TABLE 3

Degree of Amino Acid Identity and Similarity of PpPK-7 and Other Homologous Proteins GCG Gap program was used: gap penalty: 10; gap extension penalty: 0.1; score matrix: blosum62)					
Swiss-Prot #					
	P25859	O49120	Q38774	P93321	Q9ZV14
Protein name	CELL DIVISION CONTROL PROTEIN 2 HOMOLOG B	CYCLIN-DEPENDENT KINASE 1	CELL DIVISION CONTROL PROTEIN 2 HOMOLOG C	CDC2 KINASE HOMOLOG CDC2MSD	PUTATIVE SERINE/THREONINE PROTEIN KINASE
Species	<i>A. thaliana</i>	<i>Dunaliella tertiolecta</i>	<i>Antirrhinum majus</i> (Garden snapdragon)	<i>Medicago sativa</i> (Alfalfa)	<i>A. thaliana</i>
Identity %	70%	68%	70%	69%	69%
Similarity %	79%	76%	81%	79%	77%

TABLE 4

Degree of Amino Acid Identity and Similarity of PpPK-8 and Other Homologous Proteins GCG Gap program was used: gap penalty: 10; gap extension penalty: 0.1; score matrix: blosum62)					
Swiss-Prot #					
	O82754	Q9M085	Q02779	Q05609	Q39886
Protein name	PUTATIVE SERINE/THREONINE KINASE	PROTEIN KINASE-LIKE PROTEIN	MITOGEN-ACTIVATED PROTEIN KINASE KINASE 10	SERINE/THREONINE-PROTEIN KINASE CTR1	PROTEIN KINASE
Species	<i>A. thaliana</i>	<i>A. thaliana</i>	<i>Homo sapiens</i> (Human)	<i>A. thaliana</i>	<i>Glycine max</i> (Soybean)
Identity %	25%	26%	27%	27%	26%
Similarity %	42%	40%	38%	40%	40%

TABLE 5

Degree of Amino Acid Identity and Similarity of PpPK-9 and Other Homologous Proteins GCG Gap program was used: gap penalty: 10; gap extension penalty: 0.1; score matrix: blosum62)					
Swiss-Prot #					
	Q9SL77	P34099	Q9TXB8	P40376	Q9SXP9
Protein name	PUTATIVE CAMP-DEPENDENT PROTEIN KINASE	CAMP-DEPENDENT PROTEIN KINASE CATALYTIC SUBUNIT	SERINE/THREONINE PROTEIN KINASE	CAMP-DEPENDENT PROTEIN KINASE CATALYTIC SUBUNIT	CAMP-DEPENDENT PROTEIN KINASE CATALYTIC SUBUNIT
Species	<i>A. thaliana</i>	<i>Dictyostelium discoideum</i> (Slime mold)	<i>Dictyostelium</i>	<i>Schizosaccharomyces pombe</i> (Fission yeast)	<i>Euglena gracilis</i>
Identity %	45%	33%	32%	33%	28%
Similarity %	60%	48%	48%	50%	40%

TABLE 6

Degree of Amino Acid Identity and Similarity of PpCK-1 and Other Homologous Proteins GCG Gap program was used: gap penalty: 10; gap extension penalty: 0.1; score matrix: blosum62)					
Swiss-Prot #					
	Q9SZI1	Q9ZUP4	P42158	Q9LW62	Q39050
Protein name	COL-0 CASEIN KINASE I-LIKE PROTEIN	PUTATIVE CASEIN KINASE I	CASEIN KINASE I, DELTA ISOFORM LIKE	CASEIN KINASE	CASEIN KINASE I
Species	<i>A. thaliana</i>	<i>A. thaliana</i>	<i>A. thaliana</i>	<i>A. thaliana</i>	<i>A. thaliana</i>
Identity %	49%	48%	48%	46%	40%
Similarity %	62%	61%	61%	58%	52%

TABLE 7

Degree of Amino Acid Identity and Similarity of PpCK-2 and Other Homologous Proteins GCG Gap program was used: gap penalty: 10; gap extension penalty: 0.1; score matrix: blosum62)					
Swiss-Prot #					
	Q9SZI1	P42158	Q9ZWB3	Q9ZUP4	Q9LSX4
Protein name	COL-0 CASEIN KINASE I-LIKE PROTEIN	CASEIN KINASE I	ADK1	PUTATIVE CASEIN KINASE I	CASEIN KINASE I

TABLE 7-continued

Degree of Amino Acid Identity and Similarity of PpCK-2 and Other Homologous Proteins GCG Gap program was used: gap penalty: 10; gap extension penalty: 0.1; score matrix: blosum62)					
Swiss-Prot #					
	Q9SZ11	P42158	Q9ZWB3	Q9ZUP4	Q9LSX4
Species	<i>A. thaliana</i> .	<i>A. thaliana</i>	<i>A. thaliana</i>	<i>A. thaliana</i>	<i>A. thaliana</i>
Identity %	64%	59%	60%	58%	57%
Similarity %	73%	66%	72%	67%	69%

TABLE 8

Degree of Amino Acid Identity and Similarity of PpCK-3 and Other Homologous Proteins GCG Gap program was used: gap penalty: 10; gap extension penalty: 0.1; score matrix: blosum62)					
Swiss-Prot #					
	O64816	Q9ZR52	P28523	Q9SN18	Q08466
Protein name	PUTATIVE CASEIN KINASE II CATALYTIC SUBUNIT	CASEIN KINASE II ALPHA SUBUNIT	CASEIN KINASE II, ALPHA CHAIN	CASEIN KINASE II, ALPHA CHAIN 2 (CK II)	CASEIN KINASE II, ALPHA CHAIN 2
Species	<i>A. thaliana</i>	<i>Zea mays</i> (Maize)	<i>Z. mays</i>	<i>A. thaliana</i>	<i>A. thaliana</i>
Identity %	87%	89%	89%	88%	88%
Similarity %	93%	94%	93%	93%	93%

TABLE 9

Degree of Amino Acid Identity and Similarity of PpMPK-2 and Other Homologous Proteins GCG Gap program was used: gap penalty: 10; gap extension penalty: 0.1; score matrix: blosum62)					
Swiss-Prot #					
	Q9M136	Q40531	Q39024	Q40353	Q07176
Protein name	MAP KINASE 4	MITOGEN-ACTIVATED PROTEIN KINASE HOMOLOG NTF6	MITOGEN-ACTIVATED PROTEIN KINASE HOMOLOG 4	MITOGEN-ACTIVATED PROTEIN KINASE HOMOLOG MMK2	MITOGEN-ACTIVATED PROTEIN KINASE HOMOLOG MMK1
Species	<i>A. thaliana</i>	<i>Nicotiana tabacum</i> (Common tobacco)	<i>A. thaliana</i>	<i>M. sativa</i>	<i>M. sativa</i>
Identity %	70%	69%	69%	68%	66%
Similarity %	80%	78%	80%	79%	76%

TABLE 10

Degree of Amino Acid Identity and Similarity of PpMPK-3 and Other Homologous Proteins GCG Gap program was used: gap penalty: 10; gap extension penalty: 0.1; score matrix: blosum62)					
Swiss-Prot #					
	Q9SUX2	P13983	Q41192	O70495	Q9RLD9
Protein name	EXTENSIN-LIKE PROTEIN	EXTENSIN	NAPRP3	PLENTY-OF-PROLINES-101	FERULOYL-COA SYNTHETASE
Species	<i>A. thaliana</i>	<i>N. tabacum</i>	<i>Nicotiana alata</i> (Winged tobacco) (Persian tobacco)	<i>Mus musculus</i> (Mouse)	<i>Pseudomonas</i> sp.



TABLE 10-continued

Degree of Amino Acid Identity and Similarity of PpMPK-3 and  
Other Homologous Proteins GCG Gap program was used: gap penalty:  
10; gap extension penalty: 0.1; score matrix: blosum62)

	Swiss-Prot #				
	Q9SUX2	P13983	Q41192	O70495	Q9RLD9
Identity %	12%	15%	22%	18%	11%
Similarity %	21%	22%	30%	26%	20%

TABLE 11

Degree of Amino Acid Identity and Similarity of PpMPK-4 and  
Other Homologous Proteins GCG Gap program was used: gap penalty:  
10; gap extension penalty: 0.1; score matrix: blosum62)

	Swiss-Prot #				
	O49975	O48616	Q9M6Q9	O80395	Q9S7U9
Protein name	PROTEIN KINASE ZMMEK1	MAP KINASE KINASE	MAP KINASE KINASE	MAP KINASE KINASE 2	MAP2K BETA PROTEIN
Species	<i>Z. mays</i>	<i>Lycopersicon esculentum</i> (Tomato)	<i>N. tabacum</i>	<i>A. thaliana</i>	<i>A. thaliana</i>
Identity %	59%	54%	53%	50%	50%

TABLE 12

Degree of Amino Acid Identity and Similarity of PpMPK-5 and  
Other Homologous Proteins GCG Gap program was used: gap penalty:  
10; gap extension penalty: 0.1; score matrix: blosum62)

	Swiss-Prot #				
	O49975	O48616	Q9M6Q9	O80395	Q9S7U9
Protein name	PROTEIN KINASE ZMMEK1	MAP KINASE KINASE	MAP KINASE KINASE	MAP KINASE KINASE 2	MAP2K BETA PROTEIN
Species	<i>Z. mays</i>	<i>L. esculentum</i>	<i>N. tabacum</i>	<i>A. thaliana</i>	<i>A. thaliana</i>
Identity %	59%	54%	53%	50%	50%
Similarity %	72%	66%	66%	62%	62%

TABLE 13

Degree of Amino Acid Identity and Similarity of PpCPK-1 and  
Other Homologous Proteins GCG Gap program was used: gap penalty:  
10; gap extension penalty: 0.1; score matrix: blosum62)

	Swiss-Prot #				
	Q9SCS2	O04290	P53681	P93520	Q41792
Protein name	CDPK- RELATED PROTEIN KINASE	CDPK- RELATED PROTEIN KINASE	CDPK- RELATED PROTEIN KINASE	CALCIUM/CAL MODULIN- DEPENDENT PROTEIN KINASE HOMOLOG	CDPK- RELATED PROTEIN KINASE

TABLE 13-continued

Degree of Amino Acid Identity and Similarity of PpCPK-1 and Other Homologous Proteins GCG Gap program was used: gap penalty: 10; gap extension penalty: 0.1; score matrix: blosum62)					
Swiss-Prot #					
	Q9SCS2	O04290	P53681	P93520	Q41792
Species	<i>A. thaliana</i>	<i>A. thaliana</i>	<i>Daucus carota</i> (Carrot)	<i>Z. mays</i>	<i>Z. mays</i>
Identity %	64%	64%	63%	63%	63%
Similarity %	76%	76%	75%	73%	74%

TABLE 14

Degree of Amino Acid Identity and Similarity of PpCPK-2 and Other Homologous Proteins GCG Gap program was used: gap penalty: 10; gap extension penalty: 0.1; score matrix: blosum62)					
Swiss-Prot #					
	Q9S7Z4	Q42479	Q41790	O81390	Q9ZPM0
Protein name	CALCIUM- DEPENDENT PROTEIN KINASE	CALCIUM- DEPENDENT PROTEIN KINASE	CALCIUM- DEPENDENT PROTEIN KINASE	CALCIUM- DEPENDENT PROTEIN KINASE	CA2+- DEPENDENT PROTEIN KINASE
Species	<i>Marchantia polymorpha</i> (Liverwort)	<i>A. thaliana</i>	<i>Z. mays</i>	<i>N. tabacum</i>	<i>Mesembryan- themum crystallinum</i> (Common ice plant)
Identity %	66%	62%	59%	59%	59%
Similarity %	75%	73%	70%	68%	70%

## Example 6

Cloning of the Full-Length *Physcomitrella Patens* cDNA Encoding for PK-6, PK-7, PK-8, PK-9, CK-1, CK-2, CK-3, MPK-2, MPK-3, MPK-4, MPK-5, CPK-1 and CPK-2

To isolate the clones encoding PK-6 (SEQ ID NO:14), PK-7 (SEQ ID NO:15), PK-8 (SEQ ID NO:16), PK-9 (SEQ ID NO:17), CK-1 (SEQ ID NO:18), CK-2 (SEQ ID NO:19), CK-3 (SEQ ID NO:20), MPK-2 (SEQ ID NO:21), MPK-3 (SEQ ID NO:22), MPK-4 (SEQ ID NO:23), MPK-5 (SEQ ID NO:24), CPK-1 (SEQ ID NO:25) and CPK-2 (SEQ ID NO:26) from *Physcomitrella patens*, cDNA libraries were created with SMART RACE cDNA Amplification kit (Clontech Laboratories) following manufacturer's instructions. Total RNA isolated as described in Example 3 was used as the template. The cultures were treated prior to RNA isolation as follows: Salt Stress: 2, 6, 12, 24, 48 hours with 1-M NaCl-supplemented medium; Cold Stress: 4° C. for the same time points as for salt; Drought Stress: cultures were incubated on dry filter paper for the same time points as for salt.

## 5' RACE Protocol

The EST sequences PK-6 (SEQ ID NO:1), PK-7 (SEQ ID NO:2), PK-8 (SEQ ID NO:3), PK-9 (SEQ ID NO:4), CK-1 (SEQ ID NO:5), CK-2 (SEQ ID NO:6), CK-3 (SEQ ID NO:7), MPK-2 (SEQ ID NO:8), MPK-3 (SEQ ID NO:9), MPK-4 (SEQ ID NO:10), MPK-5 (SEQ ID NO:11), CPK-1 (SEQ ID NO:12) and CPK-2 (SEQ ID NO:13) identified from the database search as described in Example 4 were used to design oligos for RACE (see Table 15). The extended sequences for these genes were obtained by performing Rapid Amplification of cDNA Ends polymerase chain reaction (RACE PCR) using the Advantage 2 PCR kit (Clontech Laboratories) and the SMART RACE cDNA amplification

kit (Clontech Laboratories) using a Biometra T3 Thermocycler following the manufacturer's instructions. The sequences obtained from the RACE reactions corresponded to full-length coding regions of CC-2 and CC-3 and were used to design oligos for full-length cloning of the respective genes (see below full-length amplification).

## Full-Length Amplification

Full-length clones corresponding PK-6 (SEQ ID NO:14), PK-7 (SEQ ID NO:15), PK-8 (SEQ ID NO:16), PK-9 (SEQ ID NO:17), CK-1 (SEQ ID NO:18), CK-2 (SEQ ID NO:19), CK-3 (SEQ ID NO:20), MPK-2 (SEQ ID NO:21), MPK-3 (SEQ ID NO:22), MPK-4 (SEQ ID NO:23), MPK-5 (SEQ ID NO:24), CPK-1 (SEQ ID NO:25) and CPK-2 (SEQ ID NO:26) were obtained by performing polymerase chain reaction (PCR) with gene-specific primers (see Table 15) and the original EST as the template. The conditions for the reaction were standard conditions with PWO DNA polymerase (Roche). PCR was performed according to standard conditions and to manufacturer's protocols (Sambrook et al., 1989 Molecular Cloning, A Laboratory Manual, 2nd Edition. Cold Spring Harbor Laboratory Press. Cold Spring Harbor, N.Y., Biometra T3 Thermocycler). The parameters for the reaction were: five minutes at 94° C. followed by five cycles of one minute at 94° C., one minute at 50° C. and 1.5 minutes at 72° C. This was followed by twenty five cycles of one minute at 94° C., one minute at 65° C. and 1.5 minutes at 72° C.

The amplified fragments were extracted from agarose gel with a QIAquick Gel Extraction Kit (Qiagen) and ligated into the TOPO pCR 2.1 vector (Invitrogen) following manufacturer's instructions. Recombinant vectors were transformed into Top10 cells (Invitrogen) using standard conditions (Sambrook et al. 1989. Molecular Cloning, A Laboratory Manual, 2nd Edition. Cold Spring Harbor Laboratory Press. Cold Spring Harbor, N.Y.). Transformed cells were selected for on

LB agar containing 100 µg/ml carbenicillin, 0.8 mg X-gal (5-bromo-4-chloro-3-indolyl-β-D-galactoside) and 0.8 mg IPTG (isopropylthio-β-D-galactoside) grown overnight at 37° C. White colonies were selected and used to inoculate 3 ml of liquid LB containing 100 µg/ml ampicillin and grown overnight at 37° C. Plasmid DNA was extracted using the

QIAprep Spin Miniprep Kit (Qiagen) following manufacturer's instructions. Analyses of subsequent clones and restriction mapping was performed according to standard molecular biology techniques (Sambrook et al., 1989 Molecular Cloning, A Laboratory Manual, 2nd Edition. Cold Spring Harbor Laboratory Press. Cold Spring Harbor, N.Y.).

TABLE 15

Scheme and primers used for cloning of full-length clones				
Gene	Final product Sites	Isolation Method	Primers Race	Primers RT-PCR
PpPK-6	XmaI/HpaI	5' RACE and RT-PCR for Full-length clone	RC782: (SEQ ID NO: 43) CCACGGTCTTCGGC TGCTGGTCGTG RC783: (SEQ ID NO: 44) GCAGCACAGCACC ACCAGCGGCTAT NVT: (SEQ ID NO: 45) GCGCCAGTGAGTA GCTCCAGCATT	RC858: (SEQ ID NO: 46) ATCCCGGGTGAGTA TCACTTACGGTGGC GA RC859: (SEQ ID NO: 47) GCGTTAACTCGACC AAGGTCATATTCC AAGCA
PpPK-7	XmaI/HpaI	5' RACE and RT-PCR for Full-length clone	RC250: (SEQ ID NO: 48) CGGTGCCACCTCG TTCTGTGGTT	RC590: (SEQ ID NO: 49) ATCCCGGGAGTGGG TGGTGGACTGTAA GGA RC591: (SEQ ID NO: 50) GCGTTAACCTTCGTC TTGGACAGGTAGAG GTTAC
PpPK-8	XmaI/HpaI	5' RACE and RT-PCR for Full-length clone	(SEQ ID NO: 51) GACTCAGCCCCGTA ATCCTTCAACA	RC1016: (SEQ ID NO: 52) ATCCCGGGCAACGA GAAGCATTCGAGAT GGC RC1021: (SEQ ID NO: 53) GCGTTAACGAGCAT CACGATACTCGGTG ATTTC
PpPK-9	XmaI/SacI	5' RACE and RT-PCR for Full-length clone	RC263: (SEQ ID NO: 54) CGACGGCTAATACC ACGTTGGCGACCA	RC831: (SEQ ID NO: 55) ATCCCGGGCTGTGA TGTCGGTGTGGTGCT CTGC RC832: (SEQ ID NO: 56) GCGAGCTCGCACCA CTGAATGATGGAGA CTCAGG
PpCK-1	XmaI/HpaI	5' RACE and RT-PCR for Full-length clone	NVT: (SEQ ID NO: 57) CGACCGCAGCCCAT GAGGAAGTTAT	RC614: (SEQ ID NO: 58) ATCCCGGGCTCACG TAGTGCCTGAACT CTGTC RC615: (SEQ ID NO: 59) GCGTTAACATGCC ATCTTCTCATACTCA GACC

TABLE 15-continued

Scheme and primers used for cloning of full-length clones				
Gene	Final product Sites	Isolation Method	Primers Race	Primers RT-PCR
PpCK-2	XmaI/HpaI	5' RACE and RT-PCR for Full-length clone	NVT: (SEQ ID NO: 60) CTCGCTACCAAGC CCCATTAGAAA	RC1012: (SEQ ID NO: 61) ATCCCGGGTTGTGCG AGGACGGAGAGAGA AGAG RC1015: (SEQ ID NO: 62) GCGTTAACCTTAGG AATCGTATGGCAGA GAGCT
PpCK-3	HpaI/SacI	5' RACE and RT-PCR for Full-length clone	NVT: (SEQ ID NO: 63) GCTTCAATGTTG GGCCCTCCACA	RC640: (SEQ ID NO: 64) GCGTTAACGGGAGG AAGGTCGGGGGAAG AGACG RC641: (SEQ ID NO: 65) GCGAGCTCAGCGCT TCGCACAAC TGAGA AACCT
PpMPK-2	XmaI/HpaI	5' RACE and RT-PCR for Full-length clone	NVT: (SEQ ID NO: 66) ACGAGAAGTTGGT GGGCTTCAAGT	RC664: (SEQ ID NO: 67) ATCCCGGGCGAGCC ATGGCGCCACTTGCT T RC665: (SEQ ID NO: 68) GCGTTAACCGCGAG CAACAATGTCTGCT GGATG
PpMPK-3	XmaI/HpaI	5' RACE and RT-PCR for Full-length clone	RC268: (SEQ ID NO: 69) CCCGTAAGCCATC GGAGTGTGGAA	RC662: (SEQ ID NO: 70) ATCCCGGGCTTGAT TGGCTCGATAATTT RC663: (SEQ ID NO: 71) GCGTTAACGGCAAT ATCTGCACAGCCGTT CACT
PpMPK-4	XmaI/SacI	5' RACE and RT-PCR for Full-length clone	NVT: (SEQ ID NO: 72) GTGTCTCGCTGGGC CAAGGAATGAA	RC1001: (SEQ ID NO: 73) ATCCCGGGCGGTGCG AGTCTATTAGGTG TTGTTTC RC1005: (SEQ ID NO: 74) GAGCTCCGGTAGGT CCGACCTCTCAATT G
PpMPK-5	XmaI/SacI	5' RACE and RT-PCR for Full-length clone	RC266: (SEQ ID NO: 75) GACGACGCGAAGC CCGGTGTGGTTGA	RC572: (SEQ ID NO: 76) ATCCCGGAGAGGC TGATCTGATGCTACA GT RC573: (SEQ ID NO: 77) ATGAGCTCTGGCGG ATTGGCGAGGTAGT TCGAC

TABLE 15-continued

Scheme and primers used for cloning of full-length clones				
Gene	Final product Sites	Isolation Method	Primers Race	Primers RT-PCR
pCPK-1	XmaI/HpaI	5' RACE and RT-PCR for Full-length clone	RC526: (SEQ ID NO: 78) CGGCGCAACGTAGT ATGCGCTTCCA RC723N: (SEQ ID NO: 79) CGCGGTGAACAAC ACCTTGACAGGTGAC RC767: (SEQ ID NO: 80) GCTCGGGTCAGCCC TCAACACCGCA NVT: (SEQ ID NO: 81) GTAAAGCTTGTGC AGCAGTCATGC	RC817: (SEQ ID NO: 82) ATCCCGGGTGTAGG CGGGCAGGTTTCCA TGC RC818: (SEQ ID NO: 83) GCGTAAACGACAAC CGGAGTAGAACGGC AGTCCA
pCPK-2	XmaI/HpaI	5' RACE and RT-PCR for Full-length clone	NVT: (SEQ ID NO: 84) AGAAGCGAGGAAT GGGACGGGACGA	RC703: (SEQ ID NO: 85) ATCCCGGGCGAACT GCGATCTGAGATT CAAC RC704: (SEQ ID NO: 86) GCGTAAACGAGATC CAACCGAAGCCATC CTACGA

## Example 7

Engineering Stress-Tolerant *Arabidopsis* Plants by Over-Expressing the Genes PK-6, PK-7, PK-8, PK-9, CK-1, CK-2, CK-3, MPK-2, MPK-3, MPK-4, MPK-5, CPK-1 and CPK-2 Binary Vector Construction: Kanamycin

The plasmid construct pACGH101 was digested with PstI (Roche) and FseI (NEB) according to manufacturers' instructions. The fragment was purified by agarose gel and extracted via the Qiaex II DNA Extraction kit (Qiagen). This resulted in a vector fragment with the *Arabidopsis* Actin2 promoter with internal intron and the OCS3 terminator. Primers for PCR amplification of the NPTII gene were designed as follows:

5'NPT-Pst :  
CGC-CTG-CAG-ATT-TCA-TTT-GGA-GAG-GAC-ACG  
(SEQ ID NO: 87)

3'NPT-Fse :  
CGC-GGC-CGG-CCT-CAG-AAG-AAC-TCG-TCA-AGA-AGG-CG.  
(SEQ ID NO: 88)

The 0.9 kilobase NPTII gene was amplified via PCR from pCambia 2301 plasmid DNA [94° C. 60 sec, {94° C. 60 sec, 61° C. (-0.1° C. per cycle) 60 sec, 72° C. 2 min}×25 cycles, 72° C. 10 min on Biometra T-Gradient machine], and purified via the Qiaquick PCR Extraction kit (Qiagen) as per manufacturer's instructions. The PCR DNA was then subcloned into the pCR-BluntII TOPO vector (Invitrogen) pursuant to the manufacturer's instructions (NPT-Topo construct). These ligations were transformed into Top 10 cells (Invitrogen) and grown on LB plates with 50 ug/ml kanamycin sulfate overnight at 37° C. Colonies were then used to inoculate 2 ml LB media with 50 ug/ml kanamycin sulfate and grown overnight at 37° C. Plasmid DNA was recovered using the Qiaprep Spin Miniprep kit (Qiagen) and sequenced in both the 5' and 3' directions using standard conditions. Subsequent analysis of

the sequence data using VectorNTI software revealed no PCR errors present in the NPTII gene sequence.

The NPT-Topo construct was then digested with PstI (Roche) and FseI (NEB) according to manufacturers' instructions. The 0.9 kilobase fragment was purified on agarose gel and extracted by Qiaex II DNA Extraction kit (Qiagen). The Pst/Fse insert fragment from NPT-Topo and the Pst/Fse vector fragment from pACGH101 were then ligated together using T4 DNA Ligase (Roche) following manufacturer's instructions. The ligation was then transformed into Top10 cells (Invitrogen) under standard conditions, creating pBPSSc019 construct. Colonies were selected on LB plates with 50 ug/ml kanamycin sulfate and grown overnight at 37° C. These colonies were then used to inoculate 2 ml LB media with 50 ug/ml kanamycin sulfate and grown overnight at 37° C. Plasmid DNA was recovered using the Qiaprep Spin Mini-prep kit (Qiagen) following the manufacturer's instructions.

The pBPSSC019 construct was digested with KpnI and BsaI (Roche) according to manufacturer's instructions. The fragment was purified via agarose gel and then extracted via the Qiaex II DNA Extraction kit (Qiagen) as per its instructions, resulting in a 3 kilobase Act-NPT cassette, which included the *Arabidopsis* Acting promoter with internal intron, the NPTII gene and the OCS3 terminator.

The pBPSSJH001 vector was digested with SpeI and ApaI (Roche) and blunt-end filled with Klenow enzyme and 0.1 mM dNTPs (Roche) according to manufacture's instructions. This produced a 10.1 kilobase vector fragment minus the Gentamycin cassette, which was recircularized by self-ligating with T4 DNA Ligase (Roche), and transformed into Top10 cells (Invitrogen) via standard conditions. Transformed cells were selected for on LB agar containing 50 ug/ml kanamycin sulfate and grown overnight at 37° C. Colonies were then used to inoculate 2 ml of liquid LB containing 50 ug/ml kanamycin sulfate and grown overnight at 37° C. Plasmid DNA was extracted using the QIaprep Spin Mini-

prep Kit (Qiagen) following manufacturer's instructions. The recircularized plasmid was then digested with KpnI (Roche) and extracted from agarose gel via the Qiaex II DNA Extraction kit (Qiagen) as per manufacturer's instructions.

The Act-NPT Kpn-cut insert and the Kpn-cut pBPSJH001 recircularized vector were then ligated together using T4 DNA Ligase (Roche) and transformed into Top10 cells (Invitrogen) as per manufacturers' instructions. The resulting construct, pBPSsc022, now contained the Super Promoter, the GUS gene, the NOS terminator, and the Act-NPT cassette. Transformed cells were selected for on LB agar containing 50 µg/ml kanamycin sulfate and grown overnight at 37° C. Colonies were then used to inoculate 2 ml of liquid LB containing 50 µg/ml kanamycin sulfate and grown overnight at 37° C. Plasmid DNA was extracted using the QIAprep Spin Mini-prep Kit (Qiagen) following manufacturer's instructions. After confirmation of ligation success via restriction digests, pBPSsc022 plasmid DNA was further propagated and recovered using the Plasmid Midiprep Kit (Qiagen) following the manufacturer's instructions.

Subcloning of PK-6, PK-7, PK-8, PK-9, CK-1, CK-2, CK-3, MPK-2, MPK-3, MPK-4, MPK-5, CPK-1 and CPK-2 into the Binary Vector

The fragments containing the different *Physcomitrella patens* protein kinases were subcloned from the recombinant PCR2.1 TOPO vectors by double digestion with restriction enzymes (see Table 16) according to manufacturer's instructions. The subsequence fragment was excised from agarose gel with a QIAquick Gel Extraction Kit (QIAGEN) according to manufacturer's instructions and ligated into the binary vectors pGMSG, cleaved with XmaI and Ecl136II and dephosphorylated prior to ligation. The resulting recombinant pGMSG contained the corresponding transcription factor in the sense orientation under the constitutive super promoter.

TABLE 16

Listed are the names of the various constructs of the *Physcomitrella patens* transcription factors used for plant transformation

Gene	Enzymes used to generate gene fragment	Enzymes used to restrict pBPSJH001	Binary Vector Construct
PpPK-6	XmaI/HpaI	XmaI/SacI	pBPSJyw022
PpPK-7	XmaI/HpaI	XmaI/Ecl136	pBPSJyw012
PpPK-8	XmaI/HpaI	XmaI/Ecl136	pBPSJYW030
PpPK-9	XmaI/SacI	XmaI/SacI	PBPSERG010
PpCK-1	XmaI/HpaI	XmaI/Ecl136	pBPSY012
PpCK-2	XmaI/HpaI	XmaI/Ecl136	pBPSJyw034
PpCK-3	HpaI/SacI	SmaI/SacI	pBPSY011
PpMPK-2	XmaI/HpaI	XmaI/Ecl136	pBPSY016
PpMPK-3	XmaI/HpaI	XmaI/Ecl136	pBPSJyw014
PpMPK-4	XmaI/SacI	XmaI/SacI	pBPSJyw025
PpMPK-5	XmaI/SacI	XmaI/SacI	PBPSERG009
PpCPK-1	XmaI/HpaI	XmaI/Ecl136	PBPSERG019
PpCPK-2	XmaI/HpaI	XmaI/Ecl136	pBPSJyw008

#### *Agrobacterium* Transformation

The recombinant vectors were transformed into *Agrobacterium tumefaciens* C58C1 and PMP90 according to standard conditions (Hoefgen and Willmitzer, 1990).

#### Plant Transformation

*Arabidopsis thaliana* ecotype C24 were grown and transformed according to standard conditions (Bechtold 1993, Acad. Sci. Paris. 316:1194-1199; Bent et al. 1994, Science 265:1856-1860).

#### Screening of Transformed Plants

T1 seeds were sterilized according to standard protocols (Xiong et al. 1999, Plant Molecular Biology Reporter 17:

159-170). Seeds were plated on ½ Murashige and Skoog media (MS) (Sigma-Aldrich) pH 5.7 with KOH, 0.6% agar and supplemented with 1% sucrose, 0.5 g/L 2-[N-Morpholino]ethansulfonic acid (MES) (Sigma-Aldrich), 50 µg/ml kanamycin (Sigma-Aldrich), 500 µg/ml earbenicillin (Sigma-Aldrich) and 2 µg/ml benomyl (Sigma-Aldrich). Seeds on plates were vernalized for four days at 4° C. The seeds were germinated in a climatic chamber at an air temperature of 22° C. and light intensity of 40 micromol s<sup>-1</sup> m<sup>-2</sup> (white light; Philips TL 65W/25 fluorescent tube) and 16 hours light and 8 hours dark day length cycle. Transformed seedlings were selected after 14 days and transferred to ½ MS media pH 5.7 with KOH 0.6% agar plates supplemented with 0.6% agar, 1% sucrose, 0.5 g/L MES (Sigma-Aldrich), and 2 µg/ml benomyl (Sigma-Aldrich) and allowed to recover for five-seven days.

#### Drought Tolerance Screening

T1 seedlings were transferred to dry, sterile filter paper in a petri dish and allowed to desiccate for two hours at 80% RH (relative humidity) in a Percival Growth Cabinet MLR-350H, micromole s<sup>-1</sup> m<sup>-2</sup> (white light; Philips TL 65W/25 fluorescent tube). The RH was then decreased to 60% and the seedlings were desiccated further for eight hours. Seedlings were then removed and placed on ½ MS 0.6% agar plates supplemented with 2 µg/ml benomyl (Sigma-Aldrich) and 0.5 g/L MES (Sigma-Aldrich) and scored after five days.

Under drought stress conditions, PpPK-6 over-expressing *Arabidopsis thaliana* plants showed a 95% (20 survivors from 21 stressed plants) survival rate to the stress screening; PpPK-8, 40% (2 survivors from 5 stressed plants), PpPK-9, 78% (38 survivors from 49 stressed plants), PpCK-1, 50% (5 survivors from 10 stressed plants), PpCK-2, 52% (16 survivors from 31 stressed plants), PpCK-3, 60% (3 survivors from 5 stressed plants), PpMPK-2, 100% (52 survivors from 52 stressed plants), PpMPK-3, 98% (44 survivors from 45 stressed plants), PpMPK-4, 92% (11 survivors from 12 stressed plants), PpMPK-5, 100% (9 survivors from 9 stressed plants), PpCPK-1, 60% (12 survivors from 20 stressed plants), PpCPK-2, 89% (17 survivors from 19 stressed plants), whereas the untransformed control only showed an 11% survival rate (1 survivor from 9 stressed plants). It is noteworthy that the analyses of these transgenic lines were performed with T1 plants, and therefore, the results will be better when a homozygous, strong expresser is found.

TABLE 17

Gene Name	Summary of the drought stress tests		
	Drought Stress Test		
	Number of survivors	Total number of plants	Percentage of survivors
PpPK-6	20	21	95%
PpPK-8	2	5	40%
PpPK-9	38	49	78%
PpCK-1	5	10	50%
PpCK-2	16	31	52%
PpCK-3	3	5	60%
PpMPK-2	52	52	100%
PpMPK-3	44	45	98%
PpMPK-4	11	12	92%
PpMPK-5	9	9	100%

#### Freezing Tolerance Screening

Seedlings were moved to petri dishes containing ½ MS 0.6% agar supplemented with 2% sucrose and 2 µg/ml benomyl. After four days, the seedlings were incubated at 4° C. for 1 hour and then covered with shaved ice. The seedlings

were then placed in an Environmental Specialist ES2000 Environmental Chamber and incubated for 3.5 hours beginning at -1.0° C. decreasing 1° C./ hour. The seedlings were then incubated at -5.0° C. for 24 hours and then allowed to thaw at 5° C. for 12 hours. The water was poured off and the seedlings were scored after 5 days.

Under freezing stress conditions, PpPK-7 over-expressing *Arabidopsis thaliana* plants showed a 73% (8 survivors from 11 stressed plants) survival rate to the stress screening; PpPK-9, 100% (45 survivors from 45 stressed plants), PpCK-1, 100% (14 survivors from 14 stressed plants), PpMPK-2, 68% (36 survivors from 53 stressed plants), PpMPK-3, 92% (24 survivors from 26 stressed plants), PpCPK-2, 64% (7 survivors from 11 stressed plants), whereas the untransformed control only showed a 2% survival rate (1 survivor from 48 stressed plants). It is noteworthy that the analyses of these transgenic lines were performed with T1 plants, and therefore, the results will be better when a homozygous, strong expresser is found.

TABLE 18

Summary of the freezing stress tests			
Freezing Stress Test			
Gene Name	Number of survivors	Total number of plants	Percentage of survivors
PpPK-7	8	11	73%
PpPK-9	45	45	100%
PpCK-1	14	14	100%
PpMPK-2	36	53	68%
PpMPK-3	24	26	92%
PpCPK-2	7	11	64%
Control	1	48	2%

Salt Tolerance Screening

Seedlings were transferred to filter paper soaked in 1/2 MS and placed on 1/2 MS 0.6% agar supplemented with 2 µg/ml benomyl the night before the salt tolerance screening. For the salt tolerance screening, the filter paper with the seedlings was moved to stacks of sterile filter paper, soaked in 50 mM NaCl, in a petri dish. After two hours, the filter paper with the seedlings was moved to stacks of sterile filter paper, soaked with 200 mM NaCl, in a petri dish. After two hours, the filter paper with the seedlings was moved to stacks of sterile filter paper, soaked in 600 mM NaCl, in a petri dish. After 10 hours, the seedlings were moved to petri dishes containing 1/2 MS 0.6% agar supplemented with 2 µg/ml benomyl. The seedlings were scored after 5 days.

The transgenic plants are screened for their improved salt tolerance demonstrating that transgene expression confers salt tolerance.

Example 8

Detection of the PK-6, PK-7, PK-8, PK-9, CK-1, CK-2, CK-3, MPK-2, MPK-3, MPK-4, MPK-5, CPK-1 and CPK-2 Transgenes in the Transgenic *Arabidopsis* Lines

One leaf from a wild type and a transgenic *Arabidopsis* plant was homogenized in 250 µl Hexadecyltrimethyl ammonium bromide (CTAB) buffer (2% CTAB, 1.4 M NaCl, 8 mM EDTA and 20 mM Tris pH 8.0) and 1 µl β-mercaptoethanol. The samples were incubated at 60-65° C. for 30 minutes and 250 µl of Chloroform was then added to each sample. The samples were vortexed for 3 minutes and centrifuged for 5 minutes at 18,000xg. The supernatant was taken from each sample and 150 µl isopropanol was added. The samples were

incubated at room temperature for 15 minutes, and centrifuged for 10 minutes at 18,000xg. Each pellet was washed with 70% ethanol, dried, and resuspended in 20 µl TE. 4 µl of above suspension was used in a 20 µl PCR reaction using Taq DNA polymerase (Roche Molecular Biochemicals) according to the manufacturer's instructions.

Binary vector plasmid with each gene cloned in was used as positive control, and the wild-type C24 genomic DNA was used as negative control in the PCR reactions. 10 µl PCR reaction was analyzed on 0.8% agarose-ethidium bromide gel.

PpPk-6: The primers used in the reactions are:

15 GCTGACACGCCAAGCCTCGCTAGTC (SEQ ID NO: 89)  
 GCGTTAACTCGACCAAGGTCACATATCCAAAGCA (SEQ ID NO: 90)

The PCR program was as following: 30 cycles of 1 minute at 94° C., 1 minute at 62° C. and 4 minutes at 72° C., followed by 10 minutes at 72° C. A 2.8 kb fragment was produced from the positive control and the transgenic plants.

PpPk-7: The primers used in the reactions are:

25 GCTGACACGCCAAGCCTCGCTAGTC (SEQ ID NO: 89)  
 GCGTTAACCTTCGTCTTGGACAGGTAGAGGTTAC (SEQ ID NO: 91)

30 The primers were used in the first round of reactions with the following program: 30 cycles of 1 minute at 94° C., 1 minute at 62° C. and 4 minutes at 72° C., followed by 10 minutes at 72° C. A 1.1 kb fragment was generated from the positive control and the T1 transgenic plants.

35 PpPK-8: The primers used in the reactions were:

GCTGACACGCCAAGCCTCGCTAGTC (SEQ ID NO: 89)  
 GCGTTAACGAGCATCAGTACTCGGTGATTTC (SEQ ID NO: 92)

40 The PCR program was as following: 30 cycles of 1 minute at 94° C., 1 minute at 62° C. and 4 minutes at 72° C., followed by 10 minutes at 72° C. A 1.6 kb fragment was produced from the positive control and the transgenic plants.

45 PpPK-9: The primers used in the reactions are:

GCTGACACGCCAAGCCTCGCTAGTC (SEQ ID NO: 89)  
 GCGAGCTCGCACCCTGAATGATGGAGACTCAGG (SEQ ID NO: 93)

50 The PCR program was as following: 30 cycles of 1 minute at 94° C., 1 minute at 62° C. and 4 minutes at 72° C., followed by 10 minutes at 72° C. A 1.4 kb fragment was produced from the positive control and the transgenic plants.

55 PpCK-1: The primers used in the reactions are:

GCTGACACGCCAAGCCTCGCTAGTC (SEQ ID NO: 89)  
 GCGTTAACATGCCCATCTTCTCATACTCAGACC (SEQ ID NO: 94)

65 The PCR program was as following: 30 cycles of 1 minute at 94° C., 1 minute at 62° C. and 4 minutes at 72° C., followed by 10 minutes at 72° C. A 1.7 kb fragment was produced from the positive control and the transgenic plants.

57

PpCK-2: The primers used in the reactions are:

(SEQ ID NO: 89)  
GCTGACACGCCAAGCCTCGCTAGTC

(SEQ ID NO: 95)  
GCGTTAACCTTAGGAATCGTATGGCAGAGAGCT

The PCR program was as following: 30 cycles of 1 minute at 94° C., 1 minute at 62° C. and 4 minutes at 72° C., followed by 10 minutes at 72° C. A 1.9 kb fragment was produced from the positive control and the transgenic plants.

PpCK-3: The primers used in the reactions are:

(SEQ ID NO: 89)  
GCTGACACGCCAAGCCTCGCTAGTC

(SEQ ID NO: 96)  
GCGAGCTCAGCGCTTCGCACAACCTGAGAAACCT

The PCR program was as following: 30 cycles of 1 minute at 94° C., 1 minute at 62° C. and 4 minutes at 72° C., followed by 10 minutes at 72° C. A 1.2 kb fragment was produced from the positive control and the transgenic plants.

PpMPK-2: The primers used in the reactions are:

(SEQ ID NO: 89)  
GCTGACACGCCAAGCCTCGCTAGTC

(SEQ ID NO: 97)  
GCGTTAACGGCAATATCTGCACAGCCGTTCACT

The PCR program was as following: 30 cycles of 1 minute at 94° C., 1 minute at 62° C. and 4 minutes at 72° C., followed by 10 minutes at 72° C. A 1.7 kb fragment was produced from the positive control and the transgenic plants.

PpMPK-3: The primers used in the reactions are:

(SEQ ID NO: 89)  
GCTGACACGCCAAGCCTCGCTAGTC

(SEQ ID NO: 98)  
GCGTTAACGGCAATATCTGCACAGCCGTTCACT

The PCR program was as following: 30 cycles of 1 minute at 94° C., 1 minute at 62° C. and 4 minutes at 72° C., followed by 10 minutes at 72° C. A 2.2 kb fragment was produced from the positive control and the transgenic plants.

PpMPK-4: The primers used in the reactions are:

(SEQ ID NO: 89)  
GCTGACACGCCAAGCCTCGCTAGTC

(SEQ ID NO: 99)  
GAGCTCCGGTAGGTCCGACCTCTTCAATTG

The PCR program was as following: 30 cycles of 1 minute at 94° C., 1 minute at 62° C. and 4 minutes at 72° C., followed by 10 minutes at 72° C. A 1.7 kb fragment was produced from the positive control and the transgenic plants.

PpMPK-5: The primers used in the reactions are:

(SEQ ID NO: 89)  
GCTGACACGCCAAGCCTCGCTAGTC

(SEQ ID NO: 100)  
ATGAGCTCTGGCGGATTGGCGAGGTAGTTCGAC

The PCR program was as following: 30 cycles of 1 minute at 94° C., 1 minute at 62° C. and 4 minutes at 72° C., followed by 10 minutes at 72° C. A 1.4 kb fragment was produced from the positive control and the transgenic plants.

58

PpCPK-1: The primers used in the reactions are:

(SEQ ID NO: 89)  
GCTGACACGCCAAGCCTCGCTAGTC

(SEQ ID NO: 101)  
GCGTTAACGACAACCGGAGTAGAACGGCAGTCCA

The PCR program was as following: 30 cycles of 1 minute at 94° C., 1 minute at 62° C. and 4 minutes at 72° C., followed by 10 minutes at 72° C. A 2.3 kb fragment was produced from the positive control and the transgenic plants.

PpCPK-2: The primers used in the reactions are:

(SEQ ID NO: 89)  
GCTGACACGCCAAGCCTCGCTAGTC

(SEQ ID NO: 102)  
GCGTTAACGAGATCCAACCGAAGCCATCCTACGA

The PCR program was as following: 30 cycles of 1 minute at 94° C., 1 minute at 62° C. and 4 minutes at 72° C., followed by 10 minutes at 72° C. A 2.2 kb fragment was produced from the positive control and the transgenic plants.

The transgenes were successfully amplified from the T1 transgenic lines, but not from the wild type C24. This result indicates that the T1 transgenic plants contain at least one copy of the transgenes. There was no indication of existence of either identical or very similar genes in the untransformed *Arabidopsis thaliana* control which could be amplified by this method.

## Example 9

Detection of the PK-6, PK-7, PK-8, PK-9, CK-1, CK-2, CK-3, MPK-2, MPK-3, MPK-4, MPK-5, CPK-1 and CPK-2 Transgene mRNA in Transgenic *Arabidopsis* Lines

Transgene expression was detected using RT-PCR. Total RNA was isolated from stress-treated plants using a procedure adapted from (Verwoerd et al., 1989 NAR 17:2362). Leaf samples (50-100 mg) were collected and ground to a fine powder in liquid nitrogen. Ground tissue was resuspended in 500 µl of a 80° C., 1:1 mixture, of phenol to extraction buffer (100 mM LiCl, 100 mM Tris p18, 10 mM EDTA, 1% SDS), followed by brief vortexing to mix. After the addition of 250 µl of chloroform, each sample was vortexed briefly. Samples were then centrifuged for 5 minutes at 12,000×g. The upper aqueous phase was removed to a fresh eppendorf tube. RNA was precipitated by adding 1/10<sup>th</sup> volume 3M sodium acetate and 2 volumes 95% ethanol. Samples were mixed by inversion and placed on ice for 30 minutes. RNA was pelleted by centrifugation at 12,000×g for 10 minutes. The supernatant was removed and pellets briefly air-dried. RNA sample pellets were resuspended in 10 µl DEPC treated water. To remove contaminating DNA from the samples, each was treated with RNase-free DNase (Roche) according to the manufacturer's recommendations. cDNA was synthesized from total RNA using the 1<sup>st</sup> Strand cDNA synthesis kit (Boehringer Mannheim) following manufacturer's recommendations.

PCR amplification of a gene-specific fragment from the synthesized cDNA was performed using Tag DNA polymerase (Roche) and gene-specific primers (see Table 15 for primers) in the following reaction: 1× PCR buffer, 1.5 mM MgCl<sub>2</sub>, 0.2 µM each primer, 0.2 µM dNTPs, 1 unit polymerase, 5 µl cDNA from synthesis reaction. Amplification was performed under the following conditions: Denaturation, 95° C., 1 minute; annealing, 62° C., 30 seconds; extension, 72° C., 1 minute, 35 cycles; extension, 72° C., 5 minutes; hold, 4° C., forever. PCR products were run on a 1% agarose



gel, stained with ethidium bromide, and visualized under UV light using the Quantity-One gel documentation system (Bio-Rad).

Expression of the transgenes was detected in the T1 transgenic line. This result indicated that the transgenes are expressed in the transgenic lines and strongly suggested that their gene product improved plant stress tolerance in the transgenic line. On the other hand, no expression of identical or very similar endogenous genes could be detected by this method. These results are in agreement with the data from Example 7. This greatly supports our statement that the observed stress tolerance is due to the introduced transgene.

PpPK-6 (SEQ ID NO: 103)  
 CCCAGTAATAGCAGGGTTGGAGGAA  
 (SEQ ID NO: 104)  
 GGCTGCCTGAAGATCCGCTACAGAG  
 PpPK-7 (SEQ ID NO: 105)  
 CGTCAGGCTACTTTGCGTGGAGCAC  
 (SEQ ID NO: 106)  
 CGGTGCTGGCTAACACCAGGCCAGA  
 PpPK-8 (SEQ ID NO: 107)  
 ATCCCGGGCAACGAGAAGCATTGAGATGGC  
 (SEQ ID NO: 108)  
 GCGTTAACGAGCATCACGATACTCGGTGATTTT  
 PpPK-9 (SEQ ID NO: 109)  
 CGTGGCATCTCTCCCGATGTCTTTA  
 (SEQ ID NO: 110)  
 GGCCAACTGAAGCGGTGCATGATC  
 PpCK-1 (SEQ ID NO: 111)  
 CTCGAGGGCTCGTTACCCGTGACCT  
 (SEQ ID NO: 112)  
 CGGAGGTAACAGTAGTCAGGCTGCTC  
 PpCK-2 (SEQ ID NO: 113)  
 CCGCGACCCCTCCACGCATCAGCAT  
 (SEQ ID NO: 114)  
 CCTCCAGGAAGCCTGC GCCGAGAAG  
 PpCK-3 (SEQ ID NO: 115)  
 GGACATGTCCGTGATCAGCAATCGA  
 (SEQ ID NO: 116)  
 CAGCCTCTGGAACAACCAGACGCTG  
 PpMPK-2 (SEQ ID NO: 117)  
 GTCACCGCGAGGTACAAGCCACCAC  
 (SEQ ID NO: 118)  
 GCAGCTCTGGAGTCTGTACCACCT  
 PpMPK-3 (SEQ ID NO: 119)  
 ACGGCCACGTCGAGAATCTGAGCAA  
 (SEQ ID NO: 120)  
 CGAAGTGCTCGCAAGCAATGCCGAA

-continued

PpMPK-4 (SEQ ID NO: 121)  
 ATCCCGGGCGGTCGAGTCGTATTAGGTGTTGTTTC  
 (SEQ ID NO: 122)  
 GAGCTCCGGTAGGTCCGACCTCTTCAATTG  
 PpMPK-5 (SEQ ID NO: 123)  
 GGGCAACTGTCAATAGCAGACCTGGA  
 (SEQ ID NO: 124)  
 GCAAGTCCCAACGAACGTGTCTCGCT  
 PpCPK-1 (SEQ ID NO: 125)  
 GCGAAGATGACGACTGCTATTGCGA  
 (SEQ ID NO: 126)  
 CGTGATGACTCCAATGCTCCATACG  
 PpCPK-2 (SEQ ID NO: 127)  
 GCCAGCATCGAGGTGAGTATCCGGTGT  
 (SEQ ID NO: 128)  
 GTCTGTGGCCTTCAGAGGCGCATCCTC

Amplification was performed under the following conditions: Denaturation, 95° C., 1 minute; annealing, 62° C., 30 seconds; extension, 72° C., 1 minute, 35 cycles; extension, 72° C., 5 minutes; hold, 4° C., forever. PCR products were run on a 1% agarose gel, stained with ethidium bromide, and visualized under UV light using the Quantity-One gel documentation system (Bio-Rad).

Expression of the transgenes was detected in the T1 transgenic line. These results indicated that the transgenes are expressed in the transgenic lines and strongly suggested that their gene product improved plant stress tolerance in the transgenic lines. In agreement with the previous statement, no expression of identical or very similar endogenous genes could be detected by this method. These results are in agreement with the data from Example 7.

Example 10

Engineering Stress-Tolerant Soybean Plants by Over-Expressing the PK-6, PK-7, PK-8, PK-9, CK-1, CK-2, CK-3, MPK-2, MPK-3, MPK-4, MPK-5, CPK-1 and CPK-2 Gene

The constructs pBPSJyw022, pBPSJyw012, pBPSJYW030, PBPSERG010, pBPSSY012, pBPSJyw034, pBPSSY011, pBPSSY016, pBPSJyw014, pBPSJyw025, PBPSERG009, PBPSERG019 and pBPSJyw008 are used to transform soybean as described below.

Seeds of soybean are surface sterilized with 70% ethanol for 4 minutes at room temperature with continuous shaking, followed by 20% (v/v) Clorox supplemented with 0.05% (v/v) Tween for 20 minutes with continuous shaking. Then, the seeds are rinsed 4 times with distilled water and placed on moistened sterile filter paper in a Petri dish at room temperature for 6 to 39 hours. The seed coats are peeled off, and cotyledons are detached from the embryo axis. The embryo axis is examined to make sure that the meristematic region is not damaged. The excised embryo axes are collected in a half-open sterile Petri dish and air-dried to a moisture content less than 20% (fresh weight) in a sealed Petri dish until further use.

*Agrobacterium tumefaciens* culture is prepared from a single colony in LB solid medium plus appropriate antibiotics (e.g, 100 mg/l streptomycin, 50 mg/l kanamycin) followed by growth of the single colony in liquid LB medium to an optical

density at 600 nm of 0.8. Then, the bacteria culture is pelleted at 7000 rpm for 7 minutes at room temperature, and resuspended in MS (Murashige and Skoog, 1962) medium supplemented with 100  $\mu\text{M}$  acetosyringone. Bacteria cultures are incubated in this pre-induction medium for 2 hours at room temperature before use. The axis of soybean zygotic seed embryos at approximately 15% moisture content are imbibed for 2 hours at room temperature with the pre-induced *Agrobacterium* suspension culture. The embryos are removed from the imbibition culture and are transferred to Petri dishes containing solid MS medium supplemented with 2% sucrose and incubated for 2 days, in the dark at room temperature. Alternatively, the embryos are placed on top of moistened (liquid MS medium) sterile filter paper in a Petri dish and incubated under the same conditions described above. After this period, the embryos are transferred to either solid or liquid MS medium supplemented with 500 mg/L carbenicillin or 300 mg/L cefotaxime to kill the *agrobacteria*. The liquid medium is used to moisten the sterile filter paper. The embryos are incubated during 4 weeks at 25° C., under 150  $\mu\text{mol m}^{-2}\text{sec}^{-1}$  and 12 hours photoperiod. Once the seedlings produce roots, they are transferred to sterile metromix soil. The medium of the in vitro plants is washed off before transferring the plants to soil. The plants are kept under a plastic cover for 1 week to favor the acclimatization process. Then the plants are transferred to a growth room where they are incubated at 25° C., under 150  $\mu\text{mol m}^{-2}\text{sec}^{-1}$  light intensity and 12 hours photoperiod for about 80 days,

The transgenic plants are then screened for their improved drought, salt and/or cold tolerance according to the screening method described in Example 7 to demonstrate that transgene expression confers stress tolerance.

#### Example 11

Engineering Stress-Tolerant Rapeseed/Canola Plants by Over-Expressing the PK-6, PK-7, PK-8, PK-9, CK-1, CK-2, CK-3, MPK-2, MPK-3, MPK-4, MPK-5, CPK-1 and CPK-2 Genes

The constructs pBPSJyw022, pBPSJyw012, pBPSJYW030, PBPSERG010, pBPSSY012, pBPSJyw034, pBPSSY011, pBPSSY016, pBPSJyw014, pBPSJyw025, PBPSERG009, PBPSERG019 and pBPSJyw008 are used to transform rapeseed/canola as described below.

The method of plant transformation described herein is also applicable to *Brassica* and other crops. Seeds of canola are surface sterilized with 70% ethanol for 4 minutes at room temperature with continuous shaking, followed by 20% (v/v) Clorox supplemented with 0.05% (v/v) Tween for 20 minutes, at room temperature with continuous shaking. Then, the seeds are rinsed 4 times with distilled water and placed on moistened sterile filter paper in a Petri dish at room temperature for 18 hours. Then the seed coats are removed and the seeds are air dried overnight in a half-open sterile Petri dish. During this period, the seeds lose approx. 85% of its water content. The seeds are then stored at room temperature in a sealed Petri dish until further use. DNA constructs and embryo imbibition are as described in Example 10. Samples of the primary transgenic plants (T0) are analyzed by PCR to confirm the presence of T-DNA. These results are confirmed by Southern hybridization in which DNA is electrophoresed on a 1% agarose gel and transferred to a positively charged nylon membrane (Roche Diagnostics). The PCR DIG Probe Synthesis Kit (Roche Diagnostics) is used to prepare a digoxigenin-labelled probe by PCR, and used as recommended by the manufacturer.

The transgenic plants are then screened for their improved stress tolerance according to the screening method described in Example 7 to demonstrate that transgene expression confers drought tolerance.

#### Example 12

Engineering Stress-Tolerant Plants by Over-Expressing the PK-6, PK-7, PK-8, PK-9, CK-1, CK-2, CK-3, MPK-2, MPK-3, MPK-4, MPK-5, CPK-1 and CPK-2 Genes

The constructs pBPSJyw022, pBPSJyw012, pBPSJYW030, PBPSERG010, pBPSSY012, pBPSJyw034, pBPSSY011, pBPSSY016, pBPSJyw014, pBPSJyw025, PBPSERG009, PBPSERG019 and pBPSJyw008 are used to transform corn as described below.

Transformation of maize (*Zea Mays* L.) is performed with the method described by Ishida et al. 1996. Nature Biotech 14745-50. Immature embryos are co-cultivated with *Agrobacterium tumefaciens* that carry "super binary" vectors, and transgenic plants are recovered through organogenesis. This procedure provides a transformation efficiency of between 2.5% and 20%. The transgenic plants are then screened for their improved drought, salt and/or cold tolerance according to the screening method described in Example 7 to demonstrate that transgene expression confers stress tolerance.

#### Example 13

Engineering Stress-Tolerant Wheat Plants by Over-Expressing the PK-6, PK-7, PK-8, PK-9, CK-1, CK-2, CK-3, MPK-2, MPK-3, MPK-4, MPK-5, CPK-1 and CPK-2

The constructs pBPSJyw022, pBPSJyw012, pBPSJYW030, PBPSERG010, pBPSSY012, pBPSJyw034, pBPSSY011, pBPSSY016, pBPSJyw014, pBPSJyw025, PBPSERG009, PBPSERG019 and pBPSJyw008 are used to transform wheat as described below.

Transformation of wheat is performed with the method described by Ishida et al. 1996 Nature Biotech. 14745-50. Immature embryos are co-cultivated with *Agrobacterium tumefaciens* that carry "super binary" vectors, and transgenic plants are recovered through organogenesis. This procedure provides a transformation efficiency between 2.5% and 20%. The transgenic plants are then screened for their improved stress tolerance according to the screening method described in Example 7 to demonstrate that transgene expression confers drought tolerance.

#### Example 14

Identification of Homologous and Heterologous Genes

Gene sequences can be used to identify homologous or heterologous genes from cDNA or genomic libraries. Homologous genes (e. g. full-length cDNA clones) can be isolated via nucleic acid hybridization using for example cDNA libraries. Depending on the abundance of the gene of interest, 100,000 to 1,000,000 recombinant bacteriophages are plated and transferred to nylon membranes. After denaturation with alkali, DNA is immobilized on the membrane by e. g. UV cross linking. Hybridization is carried out at high stringency conditions. In aqueous solution hybridization and washing is performed at an ionic strength of 1 M NaCl and a temperature of 68° C. Hybridization probes are generated by e. g. radioactive (<sup>32</sup>P) nick transcription labeling (High Prime, Roche, Mannheim, Germany). Signals are detected by autoradiography.

Partially homologous or heterologous genes that are related but not identical can be identified in a manner analo-

gous to the above-described procedure using low stringency hybridization and washing conditions. For aqueous hybridization, the ionic strength is normally kept at 1 M NaCl while the temperature is progressively lowered from 68 to 42° C.

Isolation of gene sequences with homologies (or sequence identity/similarity) only in a distinct domain of (for example 10-20 amino acids) can be carried out by using synthetic radio labeled oligonucleotide probes. Radio labeled oligonucleotides are prepared by phosphorylation of the 5-prime end of two complementary oligonucleotides with T4 polynucleotide kinase. The complementary oligonucleotides are annealed and ligated to form concatemers. The double stranded concatemers are then radiolabeled by, for example, nick transcription. Hybridization is normally performed at low stringency conditions using high oligonucleotide concentrations. Oligonucleotide Hybridization Solution:

6×SSC  
0.01 M sodium phosphate  
1 mM EDTA (pH 8)  
0.5% SDS  
100 µg/ml denatured salmon sperm DNA  
0.1% nonfat dried milk

During hybridization, temperature is lowered stepwise to 5-10° C. below the estimated oligonucleotide T<sub>m</sub> or down to room temperature followed by washing steps and autoradiography. Washing is performed with low stringency such as 3 washing steps using 4×SSC. Further details are described by Sambrook, J. et al. (1989), "Molecular Cloning: A Laboratory Manual", Cold Spring Harbor Laboratory Press or Ausubel, F. M. et al. (1994) "Current Protocols in Molecular Biology", John Wiley & Sons.

#### Example 15

Identification of Homologous Genes by Screening Expression Libraries with Antibodies

cDNA clones can be used to produce recombinant protein for example in *E. coli* (e.g. Qiagen QIAexpress pQE system). Recombinant proteins are then normally affinity purified via Ni—NTA affinity chromatography (Qiagen). Recombinant proteins are then used to produce specific antibodies for example by using standard techniques for rabbit immunization. Antibodies are affinity purified using a Ni—NTA column saturated with the recombinant antigen as described by Gu et al., 1994 *BioTechniques* 17:257-262. The antibody can then be used to screen expression cDNA libraries to identify homologous or heterologous genes via an immunological screening (Sambrook, J. et al. (1989), "Molecular Cloning: A Laboratory Manual", Cold Spring Harbor Laboratory Press or Ausubel, F. M. et al. (1994) "Current Protocols in Molecular Biology", John Wiley & Sons).

#### Example 16

In Vivo Mutagenesis

In vivo mutagenesis of microorganisms can be performed by passage of plasmid (or other vector) DNA through *E. coli* or other microorganisms (e.g. *Bacillus* spp. or yeasts such as *Saccharomyces cerevisiae*) which are impaired in their capabilities to maintain the integrity of their genetic information. Typical mutator strains have mutations in the genes for the DNA repair system (e.g., mutHLS, mutD, mutT, etc.; for reference, see Rupp, W. D. (1996) DNA repair mechanisms, in: *Escherichia coli* and *Salmonella*, p. 2277-2294, ASM: Washington.) Such strains are well known to those skilled in the art. The use of such strains is illustrated, for example, in Greener, A. and Callahan, M. (1994) *Strategies* 7: 32-34.

Transfer of mutated DNA molecules into plants is preferably done after selection and testing in microorganisms. Transgenic plants are generated according to various examples within the exemplification of this document.

#### Example 17

In Vitro Analysis of the Function of *Physcomitrella* Genes in Transgenic Organisms

The determination of activities and kinetic parameters of enzymes is well established in the art. Experiments to determine the activity of any given altered enzyme must be tailored to the specific activity of the wild-type enzyme, which is well within the ability of one skilled in the art. Overviews about enzymes in general, as well as specific details concerning structure, kinetics, principles, methods, applications and examples for the determination of many enzyme activities may be found, for example, in the following references: Dixon, M., and Webb, E. C., (1979) *Enzymes*. Longmans: London; Fersht, (1985) *Enzyme Structure and Mechanism*. Freeman: New York; Walsh, (1979) *Enzymatic Reaction Mechanisms*. Freeman: San Francisco; Price, N. C., Stevens, L. (1982) *Fundamentals of Enzymology*. Oxford Univ. Press: Oxford; Boyer, P. D., ed. (1983) *The Enzymes*, 3rd ed. Academic Press: New York; Bisswangcr, H., (1994) *Enzymkinetik*, 2<sup>nd</sup> ed. VCH: Weinheim (ISBN 3527300325); Bergmeyer, H. U., Bergmeyer, J., Graßl, M., eds. (1983-1986) *Methods of Enzymatic Analysis*, 3<sup>rd</sup> ed., vol. I-XII, Verlag Chemie: Weinheim; and Ullmann's *Encyclopedia of Industrial Chemistry* (1987) vol. A9, *Enzymes*. VCH: Weinheim, p. 352-363.

The activity of proteins which bind to DNA can be measured by several well-established methods, such as DNA band-shift assays (also called gel retardation assays). The effect of such proteins on the expression of other molecules can be measured using reporter gene assays (such as that described in Kolmar, H. et al. (1995) *EMBO J.* 14: 3895-3904 and references cited therein). Reporter gene test systems are well known and established for applications in both pro- and eukaryotic cells, using enzymes such as β-galactosidase, green fluorescent protein, and several others.

The determination of activity of membrane-transport proteins can be performed according to techniques such as those described in Gennis, R. B. Pores, Channels and Transporters, in *Biomembranes, Molecular Structure and Function*, pp. 85-137, 199-234 and 270-322, Springer: Heidelberg (1989).

#### Example 18

Purification of the Desired Product from Transformed Organisms

Recovery of the desired product from plant material (i.e., *Physcomitrella patens* or *Arabidopsis thaliana*), fungi, algae, ciliates, *C. glutamicum* cells, or other bacterial cells transformed with the nucleic acid sequences described herein, or the supernatant of the above-described cultures can be performed by various methods well known in the art. If the desired product is not secreted from the cells, can be harvested from the culture by low-speed centrifugation, the cells can be lysed by standard techniques, such as mechanical force or sonification. Organs of plants can be separated mechanically from other tissue or organs. Following homogenization cellular debris is removed by centrifugation, and the supernatant fraction containing the soluble proteins is retained for further purification of the desired compound. If the product is secreted from desired cells, then the cells are removed from

the culture by low-speed centrifugation, and the supernate fraction is retained for further purification.

The supernatant fraction from either purification method is subjected to chromatography with a suitable resin, in which the desired molecule is either retained on a chromatography resin while many of the impurities in the sample are not, or where the impurities are retained by the resin while the sample is not. Such chromatography steps may be repeated as necessary, using the same or different chromatography resins. One skilled in the art would be well-versed in the selection of appropriate chromatography resins and in their most efficacious application for a particular molecule to be purified. The purified product may be concentrated by filtration or ultrafiltration, and stored at a temperature at which the stability of the product is maximized.

There is a wide array of purification methods known to the art and the preceding method of purification is not meant to be limiting. Such purification techniques are described, for

example, in Bailey, J. E. & Ollis, D. F. *Biochemical Engineering Fundamentals*, McGraw-Hill: New York (1986). Additionally, the identity and purity of the isolated compounds may be assessed by techniques standard in the art. These include high-performance liquid chromatography (HPLC), spectroscopic methods, staining methods, thin layer chromatography, NIRS, enzymatic assay, or microbiologically. Such analysis methods are reviewed in: Patek et al., 1994 *Appl. Environ. Microbiol.* 60:133-140; Malakhova et al., 1996 *Biotechnologiya* 11:27-32; and Schmidt et al., 1998 *Bioprocess Engineer.* 19:67-70. *Ullmann's Encyclopedia of Industrial Chemistry*, (1996) vol. A27, VCH: Weinheim, p. 89-90, p. 521-540, p. 540-547, p. 559-566, 575-581 and p. 581-587; Michal, G. (1999) *Biochemical Pathways: An Atlas of Biochemistry and Molecular Biology*, John Wiley and Sons; Falton, A. et al. (1987) *Applications of HPLC in Biochemistry in: Laboratory Techniques in Biochemistry and Molecular Biology*, vol. 17.

## APPENDIX

Nucleotide sequence of the partial PK-6 from *Physcomitrella patens*

(SEQ ID NO: 1)

GCACGAGCTCAATCCTCATGTTTCGGACTGTGGACTAGCTGCCCTTGACCATCTGG  
 TTCTGAACGCCAGGTGTCGGCACAAATGTTGGGCTCTTTCGGTTACAGTGCCCCGTA  
 GTACGCCATGTCTGGAACCTATACCGTGAAGAGTGACGCTACAGCTTCGGTGTGT  
 AATGCTGGAGCTACTCACTGGGCGCAAGCCTTTAGACAGCTCAAGACCACGATCCG  
 AGCAATCTTTGGTACGATGGGCCACACCTCAATTGCACGACATCGACGCCCTTGAC  
 GAATGGTGGATCCGTCGTTGAAGGCCATCTACCCTGCTAAATCACTCTCCGGTTG  
 CTGATATAGTCGCCCTTTGCGTCCAGCCGGAGCCGAGTTCGACCCCGATGTCG  
 AAGTGGTGCAGGCACTGTAAGGCTGATGCAGCGTGCAGTCTGAGCAAACGCAGA  
 TCGGAGTCCGCTGTTGGGAATTGAGTGAACGAGCCATCTGAGACTTACCTTTGAG  
 AGTACTGAAGCGCCCACTAGCCTAATCGTGCATCTTTGGCCATCTCGTTTCTGAGT  
 GAACCAAAGCTGGGTATATCTTTGGTGGTTAAGCAACCATTTGTCCCAATTTGAA  
 CTTCCGCTGNGAAGGCTGTATGTTGAGAACGATGCAAAGCGTTCGCGTGGTNTG  
 CTTGAACCTTCAA

Nucleotide sequence of the partial PK-7 from *Physcomitrella patens*

(SEQ ID NO: 2)

GCACGAGCTCAATCCTCATGTTTCGGACTGTGGACTAGCTGCCCTTGACCATCTGG  
 TTCTGAACGCCAGGTGTCGGCACAAATGTTGGGCTCTTTCGGTTACAGTGCCCCGTA  
 GTACGCCATGTCTGGAACCTATACCGTGAAGAGTGACGCTACAGCTTCGGTGTGT  
 AATGCTGGAGCTACTCACTGGGCGCAAGCCTTTAGACAGCTCAAGACCACGATCCG  
 AGCAATCTTTGGTACGATGGGCCACACCTCAATTGCACGACATCGACGCCCTTGAC  
 GAATGGTGGATCCGTCGTTGAAGGCCATCTACCCTGCTAAATCACTCTCCGGTTG  
 CTGATATAGTCGCCCTTTGCGTCCAGCCGGAGCCGAGTTCGACCCCGATGTCG  
 AAGTGGTGCAGGCACTGTAAGGCTGATGCAGCGTGCAGTCTGAGCAAACGCAGA  
 TCGGAGTCCGCTGTTGGGAATTGAGTGAACGAGCCATCTGAGACTTACCTTTGAG  
 AGTACTGAAGCGCCCACTAGCCTAATCGTGCATCTTTGGCCATCTCGTTTCTGAGT  
 GAACCAAAGCTGGGTATATCTTTGGTGGTTAAGCAACCATTTGTCCCAATTTGAA  
 CTTCCGCTGNGAAGGCTGTATGTTGAGAACGATGCAAAGCGTTCGCGTGGTNTG  
 CTTGAACCTTCAA

Nucleotide sequence of the partial PK-8 from *Physcomitrella patens*

(SEQ ID NO: 3)

GCACCAGACTATGACAAGCGCACGCCCTTGACATCGCCGCGTCCCTGGATTGTGTC  
 CCTGTTGCTAAAGTCCCTGCTGCGGAAGGAGCAGAGTGAATGCAAAAGACAGGTG  
 GGGGAAATCTCCGAGAGGCGAGGCGGAGAGTGCAGGATACATGGAGATGGTAAAG  
 CTGTTGAAGGATTACGGGGCTGAGTCAACGACAGGTGCCCCGAGGGGGCCAGTTGA  
 GAGTCTGATTCAAGTTGCCCTCCGTTGCCTTCAACCGCGACTGGGAGATCGCTCC  
 GTCGGAGATTGAACCTGATACAGCGAGCTCATCGGCAAAGGCTCCTTTGGAGAGA  
 TTCGGAAGGCGCTTTGGCGCGGCACACCCGTCGCTGTAAGACAATCAGACCTTCTC  
 TGTTCCAACGACAGAATGGTATCAAGGACTTCCAGCACGAGGTGCAATTGCTCGTA  
 AAGGTTCCGACCCCAAACATTTGTCAGTTCCTCGGGCTGTTACCCGTCAAAGACCT  
 CTCATGTTAGTCAACGAGTTTCTGGCAGGGGGCGATTGTCATCAGTTGTCGAGGAG  
 CACCTAAATTTGGCTCCTGACCGCATCGTGAAGTATGCCCTCNACATAGCTCGCG  
 CATGCTTACTTACCATCGGAGCAGCCCA

Nucleotide sequence of the partial PK-9 from *Physcomitrella patens*

(SEQ ID NO: 4)

TCCAGCCCAATTTGGTTGGCCACACACAGCTGTTTATGAGTCAACCGCTTCAGGNTGA  
 ACTGAAGAAACGTAACCTCCGTACGGCTATTTTACCAAATTTCAAGCTCGTTGTCCC  
 GCCATGATCCAAATGGAAGCTCAGTTTGAACATGAAGTACATTGAACACACCTTACC  
 GCCCACCAGTCAGAAGCCAGGCCATGACCTTGTCTTGAATGATCTCGGGTCTAAG

## APPENDIX-continued

AAATCAGCCATGCCACAGACTGTGAAAGTGCCTCATCCGACATTTGCTTTGCAAAC  
CGAAATCAACCAGCTGAAGTCGCTTTCCGATCTATCATAAGAACATCGGGAGA  
GATGCCACGATATACAACGCCATCCTTGTGCAGAAAGTCGACGGCTAATACCACGTT  
GGCGACCAGAAAACGAGCTGAGTTCTCGTCTAAAGGTGACCGAAGTAGAAGTTCTA  
GAGGCCCAGCTAACACACAATTAAGAACGAGTGCCACATTTGCTACTGTCAATAGGG  
GTGGCCAAGAGATGGCCACGAAATGGGGAAGGCCCTCAGTTGCTTGAAAAGAGTTCT  
CTCCAATAGGACTTGGCCCTCCGACCCGAGTCTCTGAACCTTACGCTCTCTGGTACCTT  
TTCATGCTTATGACGTCATCTGATTTCTTGACAGACCCACACCGACATCACAGCAA  
TCGGTTGAATAGACCTGGTGCCGATTCT

Nucleotide sequence of the partial CK-1 from *Physcomitrella patens*

(SEQ ID NO: 5)

TATGCCATCTTCTCATACTCAGACCAGATCCTCTATTTC AATTACAGAAGAAAGTT  
GCTTGTGCAACGTATTGAAATCATCACCGTCATGGGCTTTCCGAGTAAAAATTCTTG  
TAATGGATAAAGTCATTTCTAGTCTGATCCATACAAGCTACCGACACATGCTAGAA  
GCCTTGATTTACACACTACACACTAGAGAGTCTACAACCTTTTCTACACTCTGCTT  
AGTTGCCTCATCTCAACTCCATAAAACCCCAATTCACAATCATGTAAGACTTGAGAG  
AGGGAACAGTAAGCAACCTTGTGCTATTTTAGTACCAGAGCAGAGGATGAACCAC  
TAGTCTCCCAACGTAAGCCCTAATTCGCCGCAACAACCTCACGACGGAACCTCCGAC  
TTGGTCAAGGGTGGACAATATGATACATTCGAAGTCGATTTTGCAATGGGACGA  
AGCAGCGGAATCTGGCTGCGCACTGATTGCAGAGAGCCATTCGGGGGAGTTGAG  
TATACACAGTCCAGTCTGACATGTTGTCGAGCTGGAAATTTTCTGAATGAAAAGAT  
CACGGAACAAGCTTCGGAGGTACAGTAGTCAGGCTGCTCGTAAAAACCTANACTTC  
CGCGCTGGTGC AAAAAGTCGGCAAAATGACTGGGATACCCATCACAAAGCTCCTC  
CCACAGTGGGGTTCATCTTGATTTTGTGTGATGACTCGTGTGCTTCTGGTCAGT  
GAGGGCTTCCCGCCCTTCCCTTGCCATGGCAAAATGCCTCTTAGAAAAGTACATAA  
AATGTAACCAAGTGAATCTATGTCTCTTCTACTGTGCTCGATTCCTCTGTGCT  
GATTCCTACTAGCGTACCGTGCCGCTCCCTGTGAAGCTTCTCTATCTCGGTAAGGGA  
TATGCCCTTCGTGTGGCCGGTCCATGTAATCTTTCGCAAGCCAAAATCTAATGA  
ACACTGGTTTCTTCGCGACCGCAGCCATGAGGAAGTTATCCGGCTTCAGGTCAC  
GGTGAACGAGCCCTCGAGAATGCAGTATTCCACCCGTC AATCATTGGTAACCGA  
GCATAATCACGGTCTTCAACGAAAACCTTAGCCACACACCTTAAAGAGGTGCAAC  
AGGTTCCGGCCCAATAGGTTAGCACCATCACATGTAAGTCTTCTGTGCTTTCCGA  
ACCATCTATGTTGGGCACTCCCTTCCACCCGCAATATGTTTACAGCGCGACT  
CGTGCAATAACTCTCGTGC

Nucleotide sequence of the partial CK-2 from *Physcomitrella patens*

(SEQ ID NO: 6)

TTTTTTTTTCCAATAGATTGCAATTACATAACTCCAAGTTATGATATGTACAGGTTA  
GCAACAAGCTAATGGCTGCAAGCAGTGAACATACTACCAAGGGAGAGATTCTCACT  
CCCTAGACTTCATCCTCGTACGTACTTGGCAAGGATTATGGTTTAGTGATAAAAAG  
CTTCACAAGCCGGCAAGCATGCTGGTTGCTTCTGCTGCAATCTAATGATATTTCCCT  
AGGAATCGTATGGCAGAGACTACCACACAAGCACTGACAAATGGTTTGTAGGTTAA  
CAAGATAGAGATCCATTCAATTCCTAAGTATGAGAGACCTGTAGTCTTAGCACCATTG  
TAGGACAGAACCACCGTTTCCCTCAATCAGGCTGTGCAAAATGTAGAGCAACTC  
TCATCAACATAACAAGAGGTTTGTATAGAAGACAGAGCCCGGCTATATAACCACAA  
GCCCTGCGCTACCTTATAACGGCTTGGATCCACTCAACAGAAAGTGAATCAACTC  
CCTTGATACCGGCTTTCGTAATCCTCAAGTTGGCAGATGGCGGTTGTGGATGGCGG  
CTAGATATCCGCTTTGGGTCGCAAGTAACTGGAGAGCTCCTCTGCATCCCTGCTGAC  
GACCGTAAGCTGGTGGGACCAAGCTTACTGCTCCCTGTTCGAGAGGAATCTACGACT  
TCTGCTGATGCCCTGAGGGCTGCTGCTAGATAGGACAGCTCGCTGGAGGAAGA  
ACCCCCCGAGTTGCATACGAAGATGTATGCATGCGCTCTGGTTCTGCACACAACAGC  
AAGAGCGAATCCTTAGCAGATTCATCAAGTCCAGGACTTTTGTGCTTAGATGAGTC  
CAAAGCATTGCGACCCCGAGCCATTGCTCCTCCAGGAAGCCTGCGCCGAGAAG  
GATCATTGGTTCCGGTGGGCCGCTGCAGGTTCTCGGCTTCTGTAGCCCAAGTCCAA  
GTGCACCACTGGTTTGCCTGCAAGACCCAGTTCGAGTTGAACTGCCACCCGAA  
ATTTGTGACTGCTGGTACTTCAGAATTGTCAGTCAAAAACGTAGTCAAAATGAAAA  
CCTGTA AAACTATTTCCAGTTTAGGCAAAACAGAAGTGGCACTGTAATAAACTGAAA  
TCATCAAACTATCACAACTATCTGTTGTTGATAGAGCATAGTAAAGTCTGCGCTT  
AGGATCAAGTCTTGATACATTACAATGCCCAAGCAAGAGTGAACCTACAAAAGTT  
ACAGTTTTCATAACCTCACGAATAAAGAGGTCACGGAAGATTCCTTTCAAATATGCA  
TAGTCCGGTTTGTATCAAAACGCAAGGACCGGCAAGTGTGGAAGTACGCTCGTGC  
GAATCTGAAGGATAATTTTACAAAAGGACCTCAATGGGCGTGGACATTTGTTTTCT  
CACTGATCTTCTCGTACTTCTGCTTCTTGGTTCCCGCTTTTCACTCTGCCCATGGAA  
GACTGCCCTCAGGAAGTACATGAGCACATATCCAAGAGATTCCAAATCATCTCGTC  
TGCTTTGCTCAATCAAGATGAGTGTGATGCTTGATACCGAGCAGTCCCTGTCA  
GATTTTGTCTCCCTGAGGAAATGCTGATGCTGGAAAGGTCGCGGTAATTTCT  
GGCAAGACAAAATCAATAATGTAGACTGGTTTGTCTGCCTACCAAGCCCACTTAG  
AAAATATCAGGCTTGTGCTCTATGAAGAAAGCTTTTCGATGCACATACTCCAC  
TCTGTTGATCAGCTGGTCAGCAAGCATGAGAACAGTCTTAAAGAGAACTTCCGGCT  
GCAGAAGTTGAAAAGTCTTCGAGACTTGGCCCAACAGATCCAGAACCAAGACAT  
TGTAAGTCTCTTATCCCGAACCATCTCTGCTC

## APPENDIX-continued

---

Nucleotide sequence of the partial CK-3 from *Physcomitrella patens*

---

(SEQ ID NO: 7)

CGGTGGGGCGCTCCCCAATATTTTATCCCCGGGCTGCAGGGAATCCGGCGACCAGT  
 NTTTGAAGTGTCAACGCCGTGAATAGTGAGCGTTGCGTTATGAAGATTTTGAAGCC  
 AGTAAAGAAAAAAGATCAAAAGAGAGATCAAGATTCGCAAAACCTTTGTGGAG  
 GGCCCAACATGTGAAGCTTCTGGACATTGTCGGTATCAGCAATCGAAGACACCCA  
 GCCTAATTTTTAGTATGTGAACAATACTGATTTCAAAGTCTCTACCCACTCTTAC  
 AGACTTTGATATCCGATACATTTATGAGTCTCAAGGCTTTGGACTATTGCCA  
 TTCTCAAGGATTATGCACAGGGATGTGAAGCCACACAACGTGATGATTGACCATG  
 AGCAGCGAAGCTTATGCTTATTGACTGGGACTTGCCTGAATTTATCATCTGGCA  
 AAGAGTATAATGTGCGTGTGGCTCTAGGTACTTCAAGGGTCTGAGCTGCTGGTTG  
 ATCTCAAGATTATGATTACTCTCTCGACATGTGGAGCTTGGGGTGCATGTTTGGCC  
 GCATGATATTTCCGAAGGAGCCATCTTTTATGGGCATGACANTTCATGATCAACTT  
 GGTGAAGATCGTAAAGTGTGGGAACCTGATGAATTGAATTCCTATCTAACAAATA  
 CCGCTAAGTGGACCCCATGGAGCACCTGGTGGGGG

---

Nucleotide sequence of the partial MPK-2 from *Physcomitrella patens*

---

(SEQ ID NO: 8)

GCACGAGGAAGTAAAGCAATGTCAATCTATAATCCAATAGTGAATCACACGGGGG  
 GGAATAAGTTGCAAAACCATACAACCGCGGGATAGCGTTGTAGCCACCTAAAGAAT  
 TGAGAGTAGGCCTTACAACTTGAGATGAAGTGTGAAGTGTACTGCACCATATCATC  
 AGGACCTAAGCTGCAATCCAGAGCCTCCCTCAAATGAGATCCCTGATAGGCTCCTC  
 CGAGATAGAGGGCTCCTCGAAGCCAAACTCGAAGGGAGATACCGAGCAGGCTCAT  
 CGTTGATGTCATGAAGTGAAGCTTAAATAAGGGTGCCTCAAGGCAGCTTCCACTGTG  
 ATTCTTTTTCGCTGGATCAAAGACCAGCATCTTTTCAACAAGATCAAGAGCAGAACA  
 TTAATGCCTCTGAACTTCTGGGTAAAGGAATAGGCGACTGTGAGGCAGGTGCTGTG  
 ATATACCGCTTAGCATTTGCTGCTTCTCAAAACCCAGATCCCTATCTTCCAGGATT  
 CCGATGAGTTCTGTAATTAGGCGGAGCTGATGCACATAGTCTCTCCAGGGAACAAC  
 CGAGATCGGTTAAGCAACTCCATGAAGATGCACCCACAGACCAAAATGTCAATAGC  
 TGCAGTGTATGCTGAACAATTCAGGAGCAGCTCTGGAGCTCTGTACCACCTCGTTAC  
 AACATACTCAGTCATGAAATCCGTTTCAGAGAGAGTGCCTGCCAAGCCAAAATCTG  
 CGATTTTCAAATCGCAATTGGCATTGACGAGAAGGTGGTGGGCTTCAAGTCCCGGT  
 GCAAGACGTTTCGCGAATGGATGTAATCAAGCCCGCAAGATTGATACAGAAAA  
 TACTGACAGTGGTCTTCTGTGAGAGCTTGATTTGAACGAATGATCTGGTGTAGGTCC  
 GTATCCATCAACTCGTATACAATGTACACGTCGTTGAAATCTCGTGC

---

Nucleotide sequence of the partial MPK-3 from *Physcomitrella patens*

---

(SEQ ID NO: 9)

CGGCACCAGCCTCGCTGGAGACCACCATCGAAGCACCTTAAGCTCGTTTTCATTCCG  
 GCATTGCTTCGAGCACTTCGACTTCTAGAAATTCATAGACCTAATGGAATCGCC  
 ACTCCCTAATCTTCCGGAGAGGCTTATCGCCGACGGCAACTGCCAAGACGAGAT  
 TACTCAGATGATACTAAAAGTGCCTGCAAGGTCCGAATTAGGAATGTATGTTTCGAA  
 GAGACAGGAATTCATCTTCGAAGAGCGCGGAGCGCGCTAAGTTTGCCTGGAAGC  
 CGGTTTTCAGAGCATCTCCGAGATGAAGCTGTCTGGAATTCACACTCCGATGG  
 CTTACCCGGATAGTGGTCTCCGCGAAGAACCCCTTACCCATCTTACCTGGCC  
 CGAAGAACATTTACCGCCACGAAGTGAAGTGTCCCGCAAGGAGCAGCTCTCCG  
 CCGAAGACGCTTCACCACTCCCGAGCCCGGCATTTGTAGCGCGACTGCGATCG  
 AAGTATCTGCTGCATCTCAGCAAGTTCAACGAAATCGAGGGCAACCGAATCTCT  
 TTTATATGGCGTAGTTTGTGTCTCCGACTGGACTCTATCTATCCCATCGAGATAAC  
 TGATTCGGTGGATAATTTCTCAAATTTTGGCTAACNCAAGAACTCAGGGCGAAT

---

Nucleotide sequence of the partial MPK-4 from *Physcomitrella patens*

---

(SEQ ID NO: 10)

GCACGAGGTTGGTGAAGTTATTGATAGTGTGTGCAATTCACAGTTTTGCTACTCC  
 GGTAGGTCGACCTCTCAATGTGAGTTTAAAACTCAAAAACATTTGAGAAAAAG  
 TGTGAAAAATCTCCGTGAGGAATTCCTTGTGCAAGACGTGAAAAAAGAAAGAA  
 AGAAGATGGAATATTGTTTTGGGTATCGAAGAAGTGTTCGATGCTGTGCAATAAGG  
 AAAGAAAAAGTGCAGGTAAACATAAAAAGCTAGCATGTTGATGATAATATAAGACC  
 CGATTAAACACTTATGGATTGTTTCATGAGTGCACGTTCTCAGCGACAAAATGGGG  
 CTCATTGAGAAAACTCCACTTTCATAAGGTTGGGAACGAGCGTTTTTTTTTTGAAG  
 ATGTTTTTTCGTCATCTGATTGATATCGTTCTCAACTTGACCACATATGACTATA  
 TAAGGAAAAAGGATGAGAAAGTGGCGGATGGCGAGGTAGTTTCGACCATGCTTTT  
 GGTAAGTCCCTGGAAGTTCAGTGGTGGATCAGGCTGTGGTGTGACAGTCTCTGC  
 ACGCCATGCGAGGCTAACTTAAAGTTACAAAATCTTGCTCAAAATGGTACTCTCTC  
 GTTGTACTTTTCAGGAACCGATGTTAAGTAAATCAGTAGTTGATGGTCTGTTCACT  
 GGGACATTTCCGGATGCAAGATTCAATAAAGAACAATAATTCGGGGGAGAATTTGT  
 CAGGGATGCGGCTGCGGGGGTGGATTAACTATACATTCATGAGGATGAAGAAA  
 TTTTGCACCCCTCTTCATTCAGCTGGTTTGTATGGGAAGGTACCAACGCACACT  
 CAAAAGAGTCAATCCTAACTCCATAGGTCAGTGTGATGATGCAATGCAAGCCCT  
 GAAGCGGTTCTGNCGACATATATGTGCAAGTCCCAACGAACTGTCTCGTGGGCA  
 AGGAATGAACCAACAGCACTGACACCAAAATCAGATATTTGACCTCACCTTGT  
 GATTGATGAGGAGGTTGGAGGGCTTTATATCAGCATGATGATGTCCTGACTGGT  
 GTAGGTATTCCAATCCCTCAGAACTGACTAGCAATGACGGCAAAATACGGCTCAG  
 GTATNTGCTTCTGGTGC

## APPENDIX-continued

---

Nucleotide sequence of the partial MPK-5 from *Physcomitrella patens*

---

(SEQ ID NO: 11)

TCCCCGGCTGAGGAATTCGGCACGAGCGTTGATCCTCACCCCTGGGAAGGACCCCT  
 GGAATTGAGTAGCGTGCAGGAGCTGCATCGATCCGGAAGAGACGATGAGTAGGAGA  
 GTGAGAAGGGGAGGTCTTCGCGTGCAGGTCGCGGTCGCGAAGCAAGAGACTCCCGTCAGCAA  
 ATTTTGGACTGCCAGTGGAACTTTCAGGATGATGATCAAGCTCAACCAACCGG  
 GCTTCGCGTCTCTTCAGAACCTAACCTTCTACGCAGACGAGTCTAGTCCCC  
 AGATGGCAACTGTCAATAGCAGACCTGGAGTTAGTGCCTTCTTAGGAAAGGGTG  
 CGGGTGAACCGGTGCAGCTTGGTCCGGCACAATGGACCAATGTCAATTATGCAC  
 TGAGGCGATACAATGAATATCAACGAAACAGTGAGGAGCAGATTGTTAGGAG  
 CTGAAAATCAACCAAGTGACGCCACAGCAGTGCCTTATATCGTGAATGCTCCAC  
 TCCTTCTACCACAACCGGCTCATATCCATGATCCTAGAGTACATGGACAGGGGCTCG  
 TTGTCCGACATTATTAAGCAACAAAGCAGATACCTGAGCCGATTTTGGCCGTCATT  
 GCTAGTC

---

Nucleotide sequence of the partial CPK-1 from *Physcomitrella patens*

---

(SEQ ID NO: 12)

GCACCAGCCGAGTCGGGCATTTTTCGTGCGGTGTGAGGGCTGACCCGAGCTTTGAA  
 GAAGCCCTTGGCCTTCCATCTCTCCCGAAGCCAAAGGATTTGCGTGAAGCGTCTCCTG  
 AATAAGGATATGCGGAAACGCATGACTGCTGCACAAGCTTTAACTCATCCATGGATT  
 CGAAGTACAACGTGAAGATACCTCTGGATATCTTAGTGTACAGACTTGTGAGGAAT  
 TATCTTCGTGCATCATCCATGAGAAAGGCTGCTTTGAAGGCCCTGTCAAAGACTTTA  
 ACCGAAGACGAGACTTTTATCTACGTACTCAATTTATGCTGCTAGAACCAGTAAC  
 AACGGTCGTGTACTTTTGAGAAATTCAGACAGGCACCTGTGAAAAATTCACAGAG  
 GCCATGAAAGAGTCACGGGTTTTTGAATCTGGAATCGATGGATGGTCTTCATTTT  
 GCACCAGCCGAGTCGGGCATTTTTCGTGCGGTGTGAGGGCTGACCCGAGCTTTGAA  
 GAAGCCCTTGGCCTTCCATCTCTCCCGAAGCCAAAGGATTTGCGTGAAGCGTCTCCTG  
 AATAAGGATATGCGGAAACGCATGACTGCTGCACAAGCTTTAACTCATCCATGGATT  
 CGAAGTACAACGTGAAGATACCTCTGGATATCTTAGTGTACAGACTTGTGAGGAAT  
 TATCTTCGTGCATCATCCATGAGAAAGGCTGCTTTGAAGGCCCTGTCAAAGACTTTA  
 ACCGAAGACGAGACTTTTATCTACGTACTCAATTTATGCTGCTAGAACCAGTAAC  
 AACGGTCGTGTACTTTTGAGAAATTCAGACAGGCACCTGTGAAAAATTCACAGAG  
 GCCATGAAAGAGTCACGGGTTTTTGAATCTGGAATCGATGGATGGTCTTCATTTT  
 AAGAAAAATGGACTTTTTCAGAGTCTGTGCAGCGCCATTAGTGTCTCCAGTTAGAA  
 G

---

Nucleotide sequence of the partial CPK-2 front *Physcomitrella patens*

---

(SEQ ID NO: 13)

GCACGAGCTCCTGCATCTCCCCCTCCTTCTCCTCCTCATCTTCTGGAGCCAGCGAA  
 TCGCATCTGAGATTCCAACCTTGAAGGGCCCTCGCGTAAGCACCGGAGCTCGTTTCT  
 TAGCCTTTTGGCCCTCGCGATATTTGTACATTGTTTCTCCTGTTTATTCGATTCCCG  
 CTCTGAAAATGTGAACGGGCTGCAAGCTTGGTTTTGAGCAACGTGGAGCATTTGAA  
 GGTGTGCGCTCGTCCCTGCCCATTCCTCGTTCCTGCTGCGCCTATGTCATGACGACG  
 TGAAGGAGAGGATTTGAGGTTTTGCAAGTGATATAATCTCCCGAGGAGATTTCT  
 GTGAGTTGATTAACCTGGATCAGCGACATGGGAACTAGTTCCGAGGGATCGAG  
 GAAGTCCACTCGGCAGGTGAATCAGGGAGTCGGGTCTCAAGACACCCGAGAGAAGA  
 ATGATAGCGTCAATCCAAGACGAGACAGGGTGGTAGCGTTGGCGCAACCAACTAT  
 GCGGAAAGCACAAAGCAGTGGTGTCTCAGGCCGAGAACGATCCACCTCTGCGCCCG  
 TGTCTGCGGAGGCCGAAAGCCAGCATCGAGGTGATATCGCGTGTTTGGGTAAGC  
 CGTGTGAGATATTCGTCAATCTTACATCTGGGACGGGAGCTTGGCCGAGGGCAGT  
 TCGGAGTGACTTACTTGTGTAAGTACAGATGACGAATGAGGCGTACCGGTGCAAG  
 AGCATGCCCAACCGAAACTGACCAAGTAAAGGAGATATCGAGGATGTTAAGCGGGA  
 GGTTCAGATTATGCATCACCTGTGCGGACACCAATATCGTGGTGTAAAGGATGT  
 GTTCGAGGACAAGCATTCCTGTCATCTTGTGATGGAGCTCTGTGAGGTGGCGAGCT  
 TTTGATCGCATATTGCCAAGGGCATTACAGTGAGCGCGCCCTGCGCATATGTG  
 CAGAGTCATCGTCAATGTGGTGCACAGATGCCACTCATTAGGGTCTTCCATCGGGA  
 TCTCAAGCCAGAGAAATTTCTGTGGCCAGCAAGGCTGAGGATGCGCCTCTGAAGGC  
 CACAGACTTCGGTCTGTCAACTTTCTTAAGCCAGGAGATGTGTTCCAGGATATTGTT  
 GGAAGTGGTATTACGTGGCCCTGAAGTTTTGAAGAGAAGTTATGGTCTCTGAGCTG  
 ATGTTTGGAGTGCAGGCGTATTGTGTACATTTCTGCTGTGTTGTTACCCCCCTTCTG  
 GGCTGAAACTGA  
 GCAGGGTATCTTTGACGCTGTGCTCAAAGGGCACATAGACTTCGAGAACGAGTCCAT  
 GGCCGAAATCTCCAACGGGGCTAAGGATTTGGTGAGGAAATGCTAAACCTTAAC  
 GTGAANAT

---

Nucleotide sequence of the full-length PK-6 from *Physcomitrella patens*

---

(SEQ ID NO: 14)

ATCCCGGTGAGTATCACTTACGTTGGCGAGGGATGGCCTTTGGGTAGGAGCTGGT  
 ATATGCGGAGTCCAACAGAAGCTTGTGCAGGACTCTTGAGTTGTGCGTGCAGGGCT  
 GAGTGC CGGAAAGGATTTTCCGACGAAGAGTCAATGTGGCGTGGCAAAACGTTT  
 GAAGAGATGGGTGTGATATGAAGGCTCCGGCTAAGCAGTCTGTTGGAGTCCGACT  
 GCTCCTGTCTCTGTAGTATCTTCTCGGTGGTGGAGCTCTGTGATAGCCAAAGTTCAG  
 ACAGATCCAGTGGATACTACAGGCTTAATTTCCATGTGGTATGACTTAAAACAGAGT  
 CAATCTCTCACGGGTGGACTCAAATGCTTCTAACCTTTGTTGGGCAGCAGTGGTAC  
 GCGTGTGATGTGATGGCTCTTCTGTCAAGGAAATCAAAAATGGAAGTCCGGGTTTTG  
 AATGGAATTTTAACTCTCGTACTTTCAAACGCTTTTAAAAGCTTCGAATTTTTG

APPENDIX-continued

ATGCTAGTAAACAACAACATCGAAGGAAATATTCTCAACAGTTTCCTACGTCTCTTA
CTCAAAATGATATTGGAACAACAATAAATGACCGGAGGTCTCCCACAGTTTGTATCAAT
TGGGCGCCTTGACAGTTCGTAACCTTGAGCAACAACAATCTGACCGGCAACATGAAC
CCCAACTATTTCATGTGATCGTGAATGTGAAAACCTTCGATGTTTCCTATAACCAA
CTTGAGGCACTCTCCCGACTCCATTCTAAACCTGGCCAAAGCTTCGTTCTTGAATT
TGCAGAACATAAATTTAATGGTAAACTTCCTCGACGATTTCTCTCGGTGAAGAATT
TGCAGACTTTCAACATTGAGAACGATCAGTTCACGGGTAATATCCATCAGGTTTAC
CCAGTAATAGCAGGGTTGGAGAAATCGTCTTACATTTCCCCACCTCCAGCCCCG
GCACACCTGCTCCAGGACTCCTTCTCTTCCAGGAACATCGAATGGATCATCGTGC
ATCTCCCTTAGGGGCGATCATTGGAATAGCCGCTGGTGGTGTGTGTGCTGTTTATT
ACTAGCACTCGGCATCTGTTGTGTTGTCGTAAGCGGTCCAGAAAGCATTTGGCGA
TCCAGAGGCCACGACAGCAGCCGAAGACCGTGGTTCACACCTCCCCCTCCGCAA
AGAGCCAGAGTATCCCAGCAAGAGCATAGACAAAAACGAAACCGCAACATCTTT
GGCAGCAGTAAGAGTGAGAAGAAAGTTCAAGCACAGAGTATTTGAGCCAGCTCC
TCTTGACAAAGGAGCAGCCGACGAAACAGTGGTGAAGGCGICTCCGCCGTCAAGG
TACTGAAGGCTCCTCCTTCAATTAAGGGTATCAGCGGCTGGGTGCTGGACATCGA
AAGCAACAATTTGGCAAGGTGAACAAGAGCAATATTGCAGCCACCCCATCTCTGTA
GCGGATCTTCAGGCAGCCAAAAAGCTTCTCCAGGATAATCTGATTGGAGAAGG
GAGCATGGGTTCGCGTGTATCGTGCAGGTTTCCCAACGGCCAGGCTCTGGCCGTGAA
GAAGATCGACAGCAGCGCTCGATGGTGCAGAAATGAGGATGACTTCTTGAGTGTAG
TAGACAGTTTGGCTCGCTGCAGCATGCTAATACGGGCTGAGCTTGTGGGTTACTGTA
TTGAAACATGACCAACCGCTTGGTGTACGAGTACGTGAGTTCGTAAGCAACCTGAAC
GAATGTCTCCATTTCTCGGGTGAACACCAAGGCCCTGTCTGGAAATGTCGCGATT
AAGATTGCTTTGGGATCCGCGCGTCTCTGGAGTACTTGCACGAAGTCTGTGCACCT
CCGCTGGTTTCCACCAACTTCAAATCTGCCAATATTCTGCTAGACGATGAGCTCAAT
CCTCATGTTTCCGACTGTGACTAGCTGCCCTGCACCATCTGGTTCGAAACGCGAG
GTGTCCGCACAAATGTTGGGCTCTTTCCGI TACAGTGCCCCGAGTACGCCATGTCT
GGAACTTATACCGTGAAGAGTGACGTCTACAGCTTCGCTGGTGTGAATGCTGGAGCTA
CTCACTGGCGCAAGTCTTTAGACAGCTCAAGACCAGGATCCGAGCAATCTTTGGTA
CGATGGGCCACACCTCAATGACGACATCGACGCCCTGCACGAATGGTGGATCCG
TCGTTGAAGGGCATCTACCTGCTAAATCACTCTCTCGGTTGCTGATATAGTCGCC
TTTGGCTTCAGCCGGAGCCGAGTTCAGACCCCGATGCTGAAAGTGGTGCAGGCAC
TTGTAAGGCTGATGACGCGTGCAGTCTGAGCAACCGAGTCCGGAGTCCCGCTGTT
GGAATTGAGTGCAAACGAGCCATCTGAGACTCACTTTGAGAGTACTGAAGCCGCA
CTAGCCTAATCGTGATCTTTGGCCATCTCGTTCGAGTGGAAACAGCTGGGTA
TATTCTTTGGTGGTTAAGCAACATTTTGTCACAATTTGAACCTCAGCTGGAGAAGGG
TCTGTAGTGTGAAAGAAAAAGCAATGCAAAAGCGTTTCGGCGGGATGTGCTTTGAGAA
CTTACAAAACCTCATCAAGACTTTGAAATCTTTGTATTGCATCGAATCTTTCAATCA
GTCTCGGGTAGGATCAGTTCCTCTGTATCGGATACCCCTTTTCATCTAACATGGGACC
CTTTAATCCAGAGGATGGAGTGTGGAAATAGTAGCACTTGGTCGAGTTAAACGC

Nucleotide sequence of the full-length PK-7 from *Physcomitrella patens*

(SEQ ID NO: 15)

ATCCCGGAGTGGTGGTGGACTGTAAGGAGCTAGCGTTTTAGAGCTACAGTGGC
GTTTGTCTGTGTAGTGAAGTGAAGTGAAGTGAAGTGAAGTGAAGTGAAGTGAAGTGAAG
TATGGCAACTATGAGAAGCTGGAGAAGGTAGGAGAGGGGACTTACGGAAAGGTGT
ATAAGCCCGTGTATAAACGCTCCGGGAGCTGGTGGCGCTCAAGAAGACTAGGTTG
GAGATGGAGGAAGAAGGCGTCCCTTCCACCCCTTTGCGCGAAGTTTCGTTGTACAAA
ATGCTCTCCACAGCATGTATATCGTACGGTACTTTGCGTGGAGCAGCTCGAGAAA
GGCAGCAAGCCCATGCTCTACTTGGTCTTTGAATATATGGACACTGATCTTAAGAAAG
TATATTGACTTGACCGGTCTGGTTCGAGCGGGAAGCCTTCGCTCCCAAAGTGGTC
CAGAGTTTCAATGATCAATTTGTGACAGGGGCTTGCCCACTGTCATGGCCACGGAGTA
ATGCACAGGGATCTGAACCCAGAAATTTGCTCGTCGACAAAGCAACCCGCTCGTCTT
AAGATTGCCGACCTTGGTCTCGGTCGGGCAATCAGAGTGCAAATGAAGAGTTACACA
CAGAGATTGTTACTCTATGTTACCAGGCTCCTGAAGTCTTCTTGGAGCGACCAC
TACTCTACTCTGATATCTGGTCTGTTGGGTGATCTTCGCTGAACTCGTCCGGA
AAATGCCGCTTTCCTACTGGAGACTCCGAACTTCAGCAGCTTCTTCACATCTTCAGGTT
GCTTGGCACCCGATGAGACAATCTGGCCTGGTGTAGCCAGCACCGTATGGCA
CGAGTTTCTCAATGGAGACCACAAGATCTGTCCCTTGCTGTTCCCGGACTCAGCGC
GGTTGGCTTAGACCTTCTCGCAAATGTTGGTATTTCGAGCCCTCAAAGAGAATCTC
TGCCAAAGCCGCTTGAGCCATACTTATTTCTGCTGATGTTGATAAGACAGCAACCTA
AACACAACAGACAATTAAGAGAACAGGTAACCTCTACCTGTCAAGACGAAGG
TTAACGC

Nucleotide sequence of the full-length PK-8 from *Physcomitrella patens*

(SEQ ID NO: 16)

ATCCCGGCAACGAGAAGCATTCGAGATGGCAGATGCGAAGGAGGAAGCTGGCGCTG
CGCACGAAATGCACCTGGGCTGTGAGGAGTAAACGACGTGGGCTGTTAAGGACCAT
TCTGAAAGAAAGACAAGCAGCTCGTGAATGCTGCGGACTATGACAAAGCCACGCGCT
TGACATCGCCCGGCTCCCTGGATTGTGTCCCTGTTGCTAAAGTCTGCTTGGCGAAG
GAGCAGAGTTGAAATGCAAAAAGACAGGTGGGGAATCTCCGAGAGGCGAGCGGGA
GAGTGCAGGATACATGGAGATGTTAAAGCTGTGAAGGATTACGGGGCTGAGTCCAC
ACGCAAGTGCCTGAGGGCCAGTTGAGAGTCTGATTACGATTGCCCCCTCCGTTGC
CTTCAACCGCAGCTGGGAGATCGTCCCGTCCGAGATGGAATGATACACGAGGC
TCATCGCAAAGGCGCCTTTGGAGAGATTCGGAAGGCGCTTTGGCGCGGCACACCC
GTCGCTGTGAAGACAATCAGACCTTCTCTGTCCAAACGACAGAATGGTCATCAAGGAC
TCCAGCAGGAGTGCATTTGCTCGTAAAGGTTCCGGCACCCAAACATTTGTCAGTTT
CTCGGGCTGTTACCCGTCAAAGACCTCTCATGTAGTACCGAGTTTCTGGCAGG



## APPENDIX-continued

GGCGATTGTCATCAGTTGCTGAGGAGCAACCCATAATTGGCTCCTGACCGCATCGTG  
 AAGTATGCCCTCGACATAGCTCGCGGCATGTCTTACCTTCACAAATCGGAGCAAGCCC  
 ATCATCCACCGCGATCTCAAACCCGAAACATCATAGTGGACGAAGAGCATGAGCT  
 GAAGGTCGCGGACTTCGGACTGAGCAAGCTGATCGAGCTAAAGCTTATGTCATGATG  
 TGTACAAGATGACGGGGGGACTGGGAGTTACAGATACATGGCCCTGAGGTCCTC  
 GAACATCAACCCACGACAAATCCGTCGACGCTGTTTTCTTGGAAATGATATTATAT  
 GAGATGTTTGAAGCGTCGCTCCGTTTGGAGACAAGGATGCATACGACGCTGCCAC  
 ACTAGTTGCTAGAGACGATAAGCGGCCAGAGATGAGAGCCCAAACGATATCCCCAC  
 AAATGAAGGCATTGATCGAGGATTGCTGGTACCCTATACCCGAAAGCGACCCCTT  
 TCGTCGAAATCGTCAAAAACTCGAGGTAATGATGAGGATTGCTTATTGAGATTGC  
 CCAAGACCGTCGTCATCTCCGCGACATCTTGCATCTTCGACGCAATCTGCAGACT  
 CGTGATTGATCGGGCCAACTTCGAGCTGATCAATCTAAGTAGTCAATGCCTTACTG  
 GTCAAATTCAGCTCCGCGGACAGATTGGCTATGGTTCAAAGTATTGGATTCTCTG  
 CTTCTCAGAGCCAGAAACGACCCCGTGCATTTCTTCTCCGACGACCATTTGCG  
 ACATGAAGCACCAGACTTTGGATGTAGAAGCATGGTCTACATGCTTTGCTGTGAGC  
 CTTGACGCTCCGAGGTTGATCTCTTTAACAGCTTCTAGCCTTTCGCAATGGCTGC  
 ATCACTTAAGAAATCACCGAGTATCGTGATGCTCGTTAACGC

Nucleotide sequence of the full-length PK-9 from *Physcomitrella patens*

(SEQ ID NO: 17)

ATCCCGGCTGTGATGTCGGTGTGGTCTCTGCAAGAAATCAGATGACGTCATAAGC  
 ATGAAAAGGTACAGAGACGTTAAAGTTTCAAGACTCGGTCGGGAGGGCCAGTCTCT  
 ATTGGAGAGAATCTTTTCAAGCAACTGAGGCTTCCCATTCGTGCGCATCTCTT  
 GGCACCCCTATTGACAGTGAACAATGGGCACTCGTCTTAAATTGTGTGTAGCTGG  
 GCCTCTAGAATCTTACTTCCGTCACCTTTAGACGAGAACTCAGCTCGTTTTCTGGT  
 GCCAACGTTGATTAGCCGTCGAATCTGTCACAAGGATGGCGTTGTATATCGTGGC  
 ATCTCTCCGATGTTCTTATGATAGATCGGAAAGGACGACTTCAGCTGGTTGATTTT  
 GGTTTGCAAAAGCAATGTCGGATGAGCGCACTTTCACAGTCTGTGGCATGGCTGATT  
 TCTTAGCACCAGATCATTCAAGGACAAGGTCATGGCCTGGCTTCTGACTGGTGGG  
 CGGTAGTGTGGTTAATGTAATGTAATGTAATGTAATGTAATGTAATGTAATGTAATG  
 GGACAAACGAGCTTGAATTTTGGTAGAATAGCCGTCGCGAGCTTACGTTTCTT  
 CAAGTTTCAAGCCGTCGAAGCGGTTGACCTCATTGACAAGCTGCTGGTGGTGGACCCAA  
 CCAAGAGACTGGGCTGTGACAGCCATGGATCGCTTCCATAAGGGAACATCTTGG  
 TTCCGAGGTATAAAGTGGGACAAAGCCTCGATTGCGAGTGTGAAGTTCTTCAGAG  
 ATCATGACACGCCTTCAGTTGGCCATAGACTTTCTTCCCGTGGATGATAGTTATCAA  
 GTTTTGTATCTCAACCCGATGAAGACGATCCACCATGGCTTGGTGGCTGGTATAG  
 CTTGATGGCTCGTAGATCCCTTCTTCAAGCATCAATGGCACAGTACCGAATGGCT  
 ATAACAGAAGATGCACATTAAGTCTCCATGAACAGATACCGTAGCGCTTAGGATT  
 TTCGGTCTGTCACAAATGACGGCTCTCTTGTGAGGTTGAAATGTTGTGTCACCCGATG  
 ATCTCTACTGGCACAACCTCCAGGCTGAATCTTAAGGCCAGCTGTTTTAGGTGAGA  
 CGTTTACCTTGGTTCGAATCAGCTCGTGTGTTAAGCGCGAGTTCGATGATGATG  
 AAATGACGGTGTCTTGAAGTCTTGAAGGCAATCAATTCGCTTATGTTGTGCTCC  
 TTCCATGTGGTCAATAGGGAAGGGAACCGCTGCACTAGTCAAGTAAACGAATGCG  
 TTCAATGTATAGCATAGCGGTAGAGGTTTCGTACGAAATGTTGTTGCACTCGGTGA  
 TTATAGCGCATTTCTCTGAACATGACGAGAAATCGTGTCTCTGAGTCTCCATCATTT  
 AGTGGTGGAGCTCGC

Nucleotide sequence of the full-length CK-1 from *Physcomitrella patens*

(SEQ ID NO: 18)

ATCCCGGCTCACGTAGTGCACCTGACTCTGTCTGAATTTTAGGGATGAGAGGTAG  
 ATTTGAAGAATACCTGGTGTCTAATTTTCTGTAAATTTTACCCTTGGAGTACTCAT  
 GGATTTGGGAGGTGATCGCATGAGAGCTCCAGAGGCAGTCTCGAGAATATCAAT  
 ATAGATCATTGGACGCTTTCACAGAGCAGCACGAGCAGTTGCAAAAGCAGCAGCAG  
 CAAGATGAGTATCAGAGAAAGAAATGAAGCTCGAGACACTGCCAAAATGTTAAG  
 CAATGCGACCGTGTCTCTTCCCTCGAAGCAGTCCGGATGGACGTAGACTACGTAC  
 AGTCCGGAATAAGTATGCTGTGGAAGGTATGGTTGGGAGTGGCCATTTGCAAGG  
 TGTATCAGGGCTCCGATTTGACGAACACGAGGTTGTGGGCATCAAGCTGGAGGAT  
 ACGAGAATGAGCAGCTCAGTTAATGCACGAGTCCGCTTGTACAACATATTGCG  
 GGTGGGAAGGGAGTGCACCAATGAGATGTTTCGAAAAGAGCAAGACTACAAT  
 GTATGTTGCTAGACCTATTGGGGCCGAACCTGTGACCTCTTTAAGGTTGTGGG  
 CTAAGGTTTTCTGTTGAAGCCGATGATTATGCTCGGTTACCAATGATTGACCGGGTG  
 GAATACGTGCATTTCTCGAGGGCTCGTTACCCTGACCTGAAGCCGGATAAATCTCTC  
 ATGGGCTGCGGTCGCAAGGAAACCAAGTGTTCATTATAGATTTTGGCTTGGCAAG  
 GAGTACATGGACCCGCAACAGGAGCATATCCCTTACCAGATAGGAAAGGCTT  
 CACAGGACCGCACGGTACGCTAGTAGGAATCAGCACAGAGGAATCGAGCACAGT  
 AGAAGAGATGACATAGAATCACTTGGTTACATTTTATGTACTTTCTAAGAGGCAAT  
 TTGCCATGGCAAGGGAAGGGCGGCAACGCTCACTGACCAAGCAACACGAGTA  
 CATGCACAACAAAATCAAGATGAACACCACCTGTGGAGGAGCTTTGTGATGGGTATC  
 CCAGTCAATTTGCCGACTTTTTCACCAACGCGCGAAGTCTAGGTTTTCACGAGCAGC  
 CTGACTACTGTTACCTCCGAAGCTTGTTCGCTGATCTTTTCATTGAGAAAAATTTCCA  
 GCTCGACCATGTTGACACTGGAATGTTGATTACTCACTCCCCAGAAATGGCTCTCT  
 GCAATCAGTGGCAGCCAGAAATTCGCTGCTTCGTTCCATTTGCAAAATCGACCTTC  
 GAATGTATCATATTGTCCACCCTTGACCAAGTCCGAGTTCGCTCGTGGGTTGTTGC  
 GCGCAATTAGGGCTTACGTTGGGAGGACTAGTGGTTTCTCTGCTCTGTTACTAA  
 AATAGCACAAGGTTGCTTACTGTTTCCCTCTCAAGTCTTACATGATTGTGAATGGG  
 GGTATTGAGGTTGAGGATGAGGCAACTAAGCAGAGTGTAGGAAAAGAGTTGTAGA  
 CTCTCTAGTGTGATGTTGTAATCAAGGCTTCTAGCATTTGTGTCGGTAGCTTGTATG

## APPENDIX-continued

GATCAGACTAGAAATGACTTTATCCATTACAAGAATTTTACTCGGAAAGCCATGA  
CGGTGATGATTTCAATACGTTGCACAAGCAACTTCTTCTGTAATTGAAATAGAGGA  
TCTGGTCTGAGTATGAGAAGATGGGCATGTTAACGC

Nucleotide sequence of the full-length CK-2 from *Physcomitrella patens*

(SEQ ID NO: 19)

ATCCCGGGTTGTCGAGGACGGAGAGAGAAGAGAGAGAGAGAGAGAGAGAGGTTG  
TTGTTTAGGGGAGGCATGCGGGAGCAGGATTGGTGTAAAGTTCGTAAGGAGAAGGG  
AGTACATGCAAGTGCCTGCTTGTGCGATATCGGACAGCTGGATTGTAAATAAGCGG  
AGAGGAGGTCGTAATCAGGGGCGTACATCGATGGAGCCGCTGTGGGAAACAAG  
TATCGGCTGGGACGGAAAATTGGGAGCGGTTCTTTGGGGAGATCTATCTTGGGACC  
AATGTTGAGACCAATGAGGAGGTCGGAATAAAGCTGGAAGCATCAAGACGAAGCA  
TCCACAATTTGCTGTACGAGTCCAAGCTCTACCGGATACACAAGGAGGAACCTGGGA  
TTCCCAATATCAGATGGTTCGGGATAGAAGGAGACTACAATGTCTTGGTCTGGATC  
TGTTGGGGCCAAGTCTCGAAGACCTTTCAACTTCTGCAGCCGGAAGTCTCTTTAA  
AGACTGTTCTCATGCTTGTGACAGCTGATCAACAGAGTGGAGTATGTCATGCGA  
AAAGCTTTCTTATAGAGACATCAAGCCTGATAATTTCTAATGGGGCTTGGTAGGC  
GAGCAACACAGGCTCATATTATTGATTTGGTCTTGCCAAGAAGTACCGCCACCTT  
CCAGCATCAGCATATTTCCCTACAGGAGAACAAAATCTGACAGGGACTGCTCGG  
TATGCAAGCATCAACACTCATCTTGGTATTGAGCAAAGCAGACGAGATGATTTGGAA  
TCTCTGGATATGTCCTCATGTACTTCTGAGAGGCGAGTCTTCCATGGCAAGGACTG  
AAAGCGGAAACCAAGAGCAGAGTACGAGAAAGTACAGTGAAGAAAATGTTCCA  
CGCCATTGAGGTCCTTTGTAATAAATTATCCTTCAAGATTGCGCTCGTACTTCCACTA  
TGCCCGGCTCTTGGCTTTGATGACAAACCCGACTATGCAATTTGAAAAGAATCTT  
CCGTGACCTCTTATTCTGAGGGTTTCAATTTGACTACGTTTGTACTGGACAATT  
CTGAAGTACCAGCAGTCAAAAATTTCCGGTGGCAGTCAACTCGACTGGGTGCTTCT  
GCAGGGCAAACCAAGTGGTGCACCTGGAACCTGGGCTACAGGAAGCCGAGACCTGCA  
GCGGCCACCGAACCAATGGATCCTTCTCGGCGCAGGCTTCTCGGAGGACAAATG  
GCTCCGGGTCGCAAAATGCTTGGACTCATTAAGCACAAAAGTCTGGACTTGTATG  
AATCTGCTAAGGATTTCTGCTCTTGTCTTGTGTGTCAGAACAGAGCGCATGCATACAT  
CTTCGTATGCAACTCGGGGGGTTCTTCTCCAGGCGAGCTGTCTTATCTAGCAGCA  
GGCCCTCAGGGGCATCAGCAGAAGTCGTAGATTCTCTCGAACAGGGAGCAGTAAG  
TTTGGTCCCACAGCTTACGGTCTCAGCAGGGATGACAGAGGAGCTTCCAGTTACT  
TCGGACCAAAGCGGATATCTAGCCGCCATCCACAACCGCATGCGCAACTTGAGG  
ATTTACGAAGCCGCTATCAAGGGAGTTGAATCACTTTCTGTTGAGGTGGATCAAAGC  
CGTTATAAGTAGGCCCAAGGCTTGTGGTTATATAGCCGGGCTGTCTCTATCAAAAC  
CCTCTGTTATGTAGATGAGAGTTGCTCTACATTTGGCAACAGCCTGATGAGGGGA  
AAACGGTGGTCTGTCTACAATGGTGTAAAGACTACAGGTCTCTATACTTAGGAA  
TGAATGGATCTCTATCTTGTACCATCAAAACCATGTCAGTGCCTTGTGGTGTAGCT  
TCTGCCATACGATTCTTAAGGTTAACGC

Nucleotide sequence of the full-length CK-3 from *Physcomitrella patens*

(SEQ ID NO: 20)

GCCTTAACGGGAGGAAGGTCGGGGGAAGAGACGCTTGAGGCTGCTGAAAGGGGATT  
CACTCAGCGTCCCACCACCTTCGTCAATCTGGCGCAGAAGATCGGAAAATCGGTCCG  
ACGGCCAGGTGTTATGTCCAAGGCCCGGTTTACACAGATGTAATGTCCAACGCTC  
GAAAGATTATTGGGACTACGAGGCCCTCACCGTCCAATGGGGGACCAAGACGATT  
ACGAGGTAGTGCCTAAGGTGGGGCGAGGGAAATACAGTGAAGTTTGTGAAGGTGTC  
AACGCCGTGAATAGTGAGCGTTGCGTTATGAAGATTTGAAGCCAGTAAAGAAAA  
AAAGATCAAAGAGAGATCAAGATTCTGCAAAACCTTTGGAGGGCCCAACATTTG  
TGAAGCTTCTGGACATTGTCGCTGATCAGCAATCGAAGACACCAGCCTAATTTTTG  
AGTATGTGAACAATACTGATTTCAAAGTGCCTACCCACTCTTACAGACTTTGATA  
TCCGATACTACATTCATGAGCTGCTCAAGGCTTTGGACTATTGCCATTTCAAGGGA  
TTATGCACAGGGATGTGAAGCCACACAACGTGATGATTGACCATGAGCAGCGGAAG  
CTTAGGCTTATTGACTGGGACTTGCCGAATTTCTATCATCTGGCAAGAGTATAAT  
TGCGTGTGCTTCTAGGTACTTCAAGGGTCTGAGCTGCTGGTTGATCTTCAAGATT  
ATGATTAATCTCTCGACATGTGGAGCTTGGGGTGCATGTTTCCCGCATGATATTT  
GGAAGGAGCCATCTTTATGGGCATGACAATTTATGATCAACTTGTGAAGATTGCTA  
AGGTGTGGGAACCTGATGAATTGAATTCCTATCTAAACAAATACCGCCTAGAGCTGG  
ACCCCATTTGGAAGCACTGGTTGGCAGGCATAGCAGGAAACCTTGGTCAAAGTTC  
ATCAATGCTGATAATCAGCGTCTGGTGTCCAGAGGCTGTGGATTTTGGATAAG  
CTTACGCTACGATCATCAAGACAGGCTGACTGCAAGGAAGCTATGGCACATCCC  
TATTTTATCCCGTGAAGGTGTCGGAGGTTAGCAACCGTCCGAGTGTGATATGAA  
TTGATATATCTATATGGGCTTTCTGTGATTACGTCCCACCCGGCTACCAGGTTTCT  
CAGTTGTGCGAAGCGCTGAGCTCG

Nucleotide sequence of full-length MPK-2 from *Physcomitrella patens*

(SEQ ID NO: 21)

ATCCCGGGCAGCCATGGCGCACTTGCTTCGGCGAATGGGACTGTTTGACTTCTTC  
GCTTCGCCCCCGCTCGCCCTCACCCCTCCTGTTCTTGTCAACAGCCTCCTCCTCCG  
TCTCTGCTCTGCTGGCTGGGTAAGTTTGGGAGTGAGGAGGACGTGGTCATGGAAGAA  
GAGCCCCCTCTTTGTAGTGGACTGTCCGTAATTTGGACTGAGCCTGCCGGCTC  
ATCGCGTTTGCCTAGATTGTGGGCGGGTGCCTGTTGAAATCTTGAACCTGCTACTGG  
TCGGAACGCTCGAATGCGACTTTGATTGAAGGTCCTGGTTGTTGCTCGGTCGGGA  
TCTTACTCAGTCTCTCAATAGGACCTCTGAAGCAGTATGGAGACTAGCAGTGGAA  
TCCGAATTTGAAAGTTATAAGTACTCCGACCTACGGAGGTCATTACGTGAATAATG  
TGTGGCGGGAAGTATTTCGAAGTACCGCGAGGTACAGCCACCACCTCGTCCGAT

## APPENDIX-continued

TGGGCGGGAGCTTATGGAATCGTCTGTTCACTCTTTGATACCGTTACGGGTGAGGA  
 GGTGGCGGTAAAAAGATTGGAAACGCCTTCGACAACAGGATCGATGCGAAGCGAA  
 CACTGCGTAAAAATAAATCCTCCGGCATATGGATCATGAAAAAGTCTGTCGCATTA  
 CAGACATCATTCGTCCTCCCACTAGGGAGAATTTCAACGACGTGACATGTATACG  
 AGTTGATGGATACGGACCTACACAGATCATTCGTTCAAATCAAGCTCTCACAGAAG  
 ACCACTGTCAGTATTTTCTGTATCAAATCTTGGGGGCTTGAAGTACATCCATTGCGG  
 GAACGTCTTGACCCGGGACTTGAAGCCACCAACCTTCTCGTCAATGCCAATTGCGA  
 TTTGAAATCGCAGATTTGGCTTGGCAGCCTCTCTGAAACGGATTTATGAC  
 TGAGTATGTTGTAACGAGGTGGTACAGAGCTCCAGAGCTGCTCCTGAATTGTTGAC  
 ATACACTGCAGCTATTGACATTTGGTCTGTGGGGTGCATTTTCATGGAGTTGCTTAA  
 CCGATCTGCGTTGTTCCCTGGGAGAGACTATGTGCATCAGCTCCGCCAATTACAGA  
 ACTCATCGGAACCTCTGAAGATAGGGATCTTGGGTTTTTGAGAAGCGACAATGTAG  
 CGGTATATCAAGCACCTGCCTCGACAGTCCGCTATTCCCTTAACCCAGAAGTTACG  
 AGGCATTAATCGTTCTGCTCTGTATCTGTGTAAGATGTGTTCTTGTATCCAGCG  
 AAAAAGATCACAGTGAAGCTGCCTTGGCGCACCTTATTAGCTTCACTTCATGAC  
 ATCAACGATGAGCTGCCTCGGTATCTCCCTCGAGTTGACTTCGAGGACCCCT  
 ATCTCGGAGGAGCATATCAAGGATCTCATTGGAGGGAGGCTCGGATTCAGCTTA  
 GGTCTGATGATATGGTGCAGTAACTTCACTTCATCTCAAGTTGTAAGGCTTACT  
 TCAATCTTTAGTGGCTACAACGCTATCCCGGCTGTATGGTTTTGCAACTTATT  
 CCCCCGTTGATACACTATTGGATATAGAATGACAAATTCGTTAGTTCTTTTCCC  
 TGGCGCTATATCTTTGTCTGCACATTTTCATCCAGCAGACATTTGTTGCTGGCGTTAAC  
 GC

Nucleotide sequence of full-length MPK-3 from *Physcomitrella patens*

(SEQ ID NO: 22)

ATCCCGGCTTGTATTGGCTCGGATAAATTTATGTTGACAATTGATTTGTGAGGCTTCG  
 TATTGAGTCAGCGAGCAGGCTGAGAGTTCGGCAGCGAAGTTACACTCGACCTGGCT  
 GAAATTTGGAATGGAAGCGGTGAAGCTTCATCTGTGATTTTGGAGGTTGTTGACT  
 GATGAGAAGAGGCTCTGAGCTGAGAATGTTGCAATTTAGGGGCACACCCGTTT  
 TTGGAGTCCCTTGCCACTTATTACAATGTTGGTTTACAAGCTCGACAGTTTCAATC  
 GAACGTAGAGTTTGTAGTCGGTTCGAGGATCTATGTATCCGCTCAGCGGAGAGAGA  
 GCCTGATGTTGCCAAGCGATCGTGTGGGATTTGACTAGAAAGAGGTGGACCGCAT  
 CAGAACTATTATTCTTCTGTGAGGGAAGGATCGAGGTTCCAATGGGTCTCACTCCGT  
 TTTCTGTGTACGGTTCAAGGTTATGTCCGGTGGTCTACCCGACGGCCACGTCG  
 AGAATCTGAGCAATCTTGTAGCGTGCACGATCTTCTTCTGGGTAATCCAGACTACT  
 ATGTCTGCGGTAGCACCCCTTACACAATCACAATCGTATGGCAGCGAAGAGGTG  
 CTCGAGTATGGGTGACCTACTTCTGTTGCGCAACGCCAAATGCCAACCTTTCTTA  
 GAACGTGAGCGAAGGTAGTACATCGAGGATCCAAAGATTTTGGCCAGATTTTCCAAA  
 CATGGGTCATGTGCGGGAGTTGCGAAGCCGACGATGGGAGCACAACAGTACG  
 GAAGGTTTTTGTATATCATTCAGTAAACGATGCAGCAGCTTGAATCCATACGAAACGA  
 GGGCCAGAGCCTCACCTCGCTGGAGACCGACCATCGAAGCACCTTAAGCTCGTTTT  
 CATTGCGATTGCTTGCAGACATTCGACTTCCTAGAATTTCAATAGACCTAATGGA  
 ATCGCCACTCCCTAATCTTCCGGAGAGGCCTTATCGCCGACGGCACTGCCAAAGA  
 CGAGATTACTCAGATGATACTAAAAGTGCAGCAAGGTCGAAATAGGAATGATG  
 TTTGCAAGAGACAGGAATTTCTATCTTGAAGAGCGCGTAGGCGCGTAAGTTTGTGCT  
 GGAAGCCGTTTTGCGAGGATCTCCGAGATGAAGCTGTATGGAATTCACACTC  
 CGATGGCTTACCGGGATAGTGGTCTCCCGAAGAACGCCTTACCCCATCCTTAC  
 TGGCCCGAAGAACATTTACCGCCACGACAAGTGAAGTGTCCCGCAAAGGAGCAGT  
 CCTCCGCGAAGAACGCTCACCACTCCCGAGCCGCTTTGTAGCGGGACTGCG  
 TCGAAGTATCTGCTGCATCTCAGCAAGTTCACGAAATCGAGGCAACGCGAAATCT  
 CTTTATATGGCGTAGTTTGTGCTCGACTGAACTCTATCTATCCCCATCGAGATA  
 ACTGCATTGTTGGATAAATTTCTCAACATTTTGTCTTTCATCCTCAAGCAGCTCC  
 TCAATGGCCAGTAAATGTTACGACATGTGCAACCTCCAATACGTAGCGTTATT  
 TGTAAACCCAGTTTCAATCGAGGATCAAGGAATGGCGCAGTAAGCACTGCTACTTTG  
 TGCTTGGTATCCCGTTGTGACGAGATGTATGTGACCGCTGCTATCAGTGGGAT  
 TTTCTTGGAGCGAGATCTTGTCTCCGAGTTTGTTCATAACGTTTTGGTTGTTAGG  
 GGCTAGAGCGTACTATCAAGCAATGAGAAGTGTGCTGGTGTGGATTGACAGCAA  
 TCTTTGGAGGATTTGCTTTCCTATGTAGAACATAGCGAGGACACTTGGCCTGGTG  
 GGCACATCCATAGAACATAGTGTCTCACTTCTGGGTTGTTCCACCATAGGATCATA  
 TGACCTTCTCATCTATTTTGGGCTTTGTTTCGAGCTCATGTACCATGACTAGCGTC  
 ACTTTGACTGCGGTGATAATCGTTTGTCAATTTAGTGGAGCTTTGTAGATGATAGAT  
 GCCATTTGTACAGTAGCTTGGATGCTGTTTACAAGATAGCGGCAGCTAGAAGCCTTA  
 AACCTTAGCTACCATGTATTATTTAAACCATATATGAAGTGAACGGCTGTGCAGATA  
 TTGCCGTTAACGC

Nucleotide sequence of full-length MPK-4 from *Physcomitrella patens*

(SEQ ID NO: 23)

ATCCCGGGCGGTGAGTCGATATTAGGTGTTGTTTTCATTGTAAGGTTTCGGAAGCACG  
 GGGCACGGCGTATATACCGTTCCTTGAACGTTGATCTCACTTTTGGAAAGACTGA  
 ATTGAGTAGCGTCCGGAAGCTGCATCGATCCGGAAGAGACGATGAGTAGGAGAGTG  
 AGAAGGGGAGGTTCTCGCGTCCGCGTCCCGAAGCAAGAGACTCCCGTCAAGCAAT  
 TTTGACTGCGAGTGGAACTTCCAGGATGATGATATCAAGCTCAACCACCCGGGT  
 TCGCGTCTCTTTCAGAACCTAACCTTCTACGAGACGAGCTAGCTCCCGAGA  
 TGGGCAACTGTCAATAGCAGACCTGGAGTTAGTCCGCTTCTTGGGAAAGGTTGCGG  
 GTGGAACCGTGCAGCTTGTCCGGCACAAATGGACCAATGTCAATTTATGCACTGAAG  
 GCGATACAAATGAATATCAACGAAACAGTGAAGGAGCAGATTGTTTCAGGAGCTGAA  
 AATCAACCAAGTGAACGACAGCAGCTGCCCCATATCGTGAAGTGTCTCCACTCCT  
 CTACCACAACGGCGTATATCCATGATCCTAGAGTACATGGACAGGGCTCGTTGTC

## APPENDIX-continued

CGACATATTAAGCAACAAAAGCAGATACCTGAGCCGTATTGGCCGTCATTGCTAG  
 TCAAGTTCTGAAGGGATTGGAATACCTACACCAAGTCAGGCACATCATACATCGTGA  
 TATAAAGCCCTCCAACCTCCTCATCAATCAAGGGTGAAGTCAAAAATATCTGATTT  
 TGGTGTCAAGTGTGTGTTGTTTCATTCCTGGCCAGCGAGACACGTTTCGTTGGGAC  
 TTGCACATATATGTCGCCAGAACGCTTCAGGGGCGTTTCTATGCATACGACAGTGA  
 CCTATGAGTTTAGGATTGACTCTTTTGGAGTGTGCGTTGGGTACCTTCCCATACAA  
 ACCAGCTGGAATGGAAGAGGOTTGGCAAAATTTCTTCATCCTCATGGAATGTATAGT  
 TAATCAACCCCGCAGCCGCATCCCTGACAAATTTCCCCCGAATTTTGTCTTTT  
 ATTGAATCCTGCATCCGGAATGTCCAGTGAACGACCATCAACTACTGATTTACTT  
 AAACATCCGTTCCGCAAAAAGTACAACGAGGAAGAGTACCATTTGAGCAAGATTTT  
 GTAACTTAAAGTTAGCCTCGCATGGCTGCGAGAGACTGTCCTACTACCAAGCCTGAT  
 CCACCACTGAACTTCAAGGGACTTTACCAAAAGCATGGTTCGAACTACCTCGCCAATC  
 CGCCACTTTCTCAATGCCTTTTCTTATATAGTCATATGTTGTTCAAGTTGAGAAGCAT  
 ATCAAAATCAGATTGACGGAATAAATCCTTCAACGCCGTTTCCCAACCTTATAGAAA  
 GTGGAGTTTTCTCAATGAGCCCAATTTGTCGCTGAGAAGCTGACGCTCATGAAACAA  
 TCCATAAGTGTGTTAATCGGGTCTTATATATCATCACCATGCTAGCTTTTATGTT  
 ACCTGCATTTTCTTTCTTATTTGACAGCATCGAACACTTCTTCGATACCAAAAC  
 AATATTTCCATCTTTCTTTCTTTTTCACGCTTTGCGACAAGGAATTTCTCACGG  
 AGATTTTCAACACTTTTCTCAAAATGTTTTAGAGTTTTTAACTGACAATGAAGAG  
 GTCGGACCTACCGGACTCGC

Nucleotide sequence of full-length MPK-5 from *Physcomitrella patens*

(SEQ ID NO: 24)

ATCCCGGAGAGGCTGATCTGATGCTACAGTTTCGTGTGCAGCTAGTCTTTAGAGAT  
 TCGGGCAACGCACCTGTGTAAGATCGGAACTTTCAAATCGGTGAGTCGATATTAG  
 GTGTTGTTTCATTGTAAGGGTTCGGAAGCACGGGGCACGGCGTATATACCCTTCCC  
 TTGAACGTTGATCTCACCTTTGGAAGACCTGAAATGAGTAGCGTCGGAAGCTGCAT  
 CGATCCGGAAGAGACGATGAGTAGGAGAGTGAAGGGGAGGTTCTCGCTCGCGG  
 TGCCGAAGCAAGAGACTCCCGTCAGCAAAATTTTACTGTCAGTGGAACTTTCCAGG  
 ATGATGATATCAAGCTCAACACACACCGGGCTTCGCTCGCTCTCTTCAGAACTAACC  
 TTCTACGCAGACGCGTCTAGCTCCCGATGGGCACTGTCATAGCAGACCTGG  
 AGTTAGTGGGTTCTTAGGAAAGGGTTCGGGTGGAACCGTGCAGCTTGTCCGGCAC  
 AAATGGACCAATGTCAATTTATGCCTGAAAGCGGATACAAATGAATATCAACGAAAC  
 AGTGAGGAAGCAGATTGTTTCAAGAGCTGAAATCAACCAAGTGACGCACAGCAGT  
 GCCCTTATATCGTGAATGCTTCCACTCTTCTACCAACCGGCGTCATATCCATGAT  
 CCTAGATACATGAGACAGGGGCTCGTTGTCGACATTTAAGCAACAAAAGCAGA  
 TACCTGAGCCGATCTGGCCGTCATGCTAGTCAAGTTCTGAAGGGATTGGAATACC  
 TACCAAGTCAAGGCACATCATACATCGTGATATAAAGCCCTCCAACCTCCTCATCA  
 ATCAAGGGTGAAGTCAAATAATCTGATTTTGGTGTCAAGTGTGTTGTTGTTTTCATT  
 CCTTGGCCAGCGAGACACGTTTCTTGGGACTTGACATATATGTCGCCAGAACGCC  
 TTCAGGGGCGTTTCTATGCATACGACAGTGCCTATGGAGTTTAGGATTGACTCTTT  
 TGAGTGTGCGTTGGGTACCTTCCATACAAACAGCTGGAATGGAAGAGGGTTGG  
 CAAAATTTCTTCACTCCTATGGAATGTATAGTTAATCAACCCCGCAGCCGATCC  
 CCTGACAAATTTCTCCCGAATTTTGTCTTTTATTGAATCCTGCATCCGGAAATGTC  
 CCAGTGAACGACCATCAACTACTGATTTACTTAAACCTCCGTTCTTGCAAAAGTACA  
 ACAGGGAAGAGTACCATTTGAGCAAGATTTTGTAACTTAAAGTTAGCCTCGCATGGC  
 GTGCAGAGACTGTCACTACCACAAGCCTGATCCACCCTGAACTTCAAGGGACTTTA  
 CAAAAGCATGGTCAACTACCTCGCAATCCGCCAGAGCTCA

Nucleotide sequence of full-length CPK-1 from *Physcomitrella patens*

(SEQ ID NO: 25)

ATCCCGGTTGAGGCGGGCAGGTTTCGATGCAATGGGGCAGTGTATGGAAAGTTT  
 GATGATGGAGGCGAAGGGGAGGATTTGTTTGGAGCGGAGAAAGTGCAGGTTTCTAG  
 GAGCCAAAGCATGGATCGTGGAGCAATAGCAACCGAGGGAGCTTCAACAATGGCG  
 GGGGGCCCTCGCTATGAGAGCCAGACGTCGTTGGGAGCAGCCATCCGTTCCCG  
 CGCATCCCTCAGCTAGTCCGCTCCCTCACACAGAGCTCCCGAGCCCTTCGACC  
 CGCGAGCGAACAATTTCAAAGGCCTTTCTCTCTCTTCCCGCGAAGCACAT  
 CAGTCCAGTCTCGTGAACCGCATGGCGGAAGCCGAAGAAGGAGGGGCGATCCC  
 TGAGGCTGTGATGGTGAAGAGCCTTGGATAAGCATTTCCGCTATCACAAAGACT  
 CGCTACTAAGTATGAGCTGGGGCATGAAGTCCGTCGCGGGCACCTCGGTACACAT  
 GTTACGCAAAAGTACGGAAGGGCAGCATAAGGGACAAGCCGTCGAGTGAAGAT  
 AATTCGAAAGCGAAGATGACGACTGCTATTCGATCGAGGACGTGGGACGAGAAG  
 TGAAAATTTTGAAGGCTCTGACGGGACACAGAATTTGGTTCGATTTACGATTTCT  
 CGGAGGACCATCTAAATGTGTACATTTGTTAAGAAATATGTGAAGGAGGTGAATAT  
 TGGATCGAATTTTGTCTCGGGGAGGAAAGTACTCGGAGGAAGACGCCAAGGTTGTT  
 GTGCGGAGATTTGAGCGTTGTTGCGTTTGTCACTGCAAGGCGTTGTTCAACCGA  
 GATCTTAAAGCCTGAGAATTTCTGTTTACCACGAAGGATGAATATGCTCAGCTTAA  
 GCCATGATTTTGGATTGTGATTTTCAATCAAAACCGATGAAAGACTGAAACGATATC  
 GTTGAAGCGCATACTACGTTGCGCCGGAGGATTTGCATAGGTTATATTTCAATGGAA  
 GCTGACGATGGAGCATTTGAGTATCAGTACATTTTGTATGTTGTTAGTGCAGCCG  
 TTTTGGCGCGGACCGAGTCCGGCATTTTTCTGTCGCTGTTGAGGGCTGACCCGAGC  
 TTTGAAGAAGCCCTTGGCTTCCATCTCTCCGAAGCAAGGATTTCTGTAAGCGT  
 CTCCTGAATAAGGATATGGGAAACCGATGACTGCTGCACAAGCTTTAACTCATCCA  
 TGGATTCGAAGTAAACAAGTGAAGATACCTCTGGATATCTTAGTGTACAGCTTGTG  
 AGGAATATCTTCTGTCATCATCATGAGAAGGCTGCTTTGAAGGCCCTGTCAAG  
 ACTTTAAACGAAGACGAGACTTTTATCTACGTAATTTATGCTGCTAGAACCA  
 AGTAACAACCGTGTGTTACTTTTGAAGATTTTCAAGCAGGACTGCTGAAAATTTCA  
 ACAGAGGCCATGAAGAGTACGGGTTTTTGAATTTCTGGAATCGATGGATGGTCTT

## APPENDIX-continued

CATTTCAGAAAATGGACTTTTCAGAGTTCGTGCAGCGGCCATTAGTGTTCTCCAG  
TTAGAAGCCAAGAACGATGGGAGCAGCATGCTCGCCGAGCTTACGACATATTTGA  
GAAAAGGGTAAACCGAGTCATTTATCCTGATGAACTTGCAGAAAGAGATGGGACTAG  
CACCAAAATGTACCAGCCCAAGTGTTCCTAGATTGGATTAGACAGTCTGATGGTCGGC  
TGAGTTTCACTGGGTTCCACCAAGCTGTACATGGAATTTCCAGCCGTGTATCAAAA  
ATCTCCAGCAGTGATTCTTTGCATCGTACAGTTCGGAATGGAGTTTTTAAGCTCTTTT  
AGTTTCACTTCCGCTTCTCAACTGCTGCTTCGCCCTCGTCTCTGAGCTGTGATAGCGTAT  
CTCAAGCATATGCACAACCTCGCATTTTGTGTAAGTGAATTTGTACCTCACATTAGTC  
GGCCCTCTGGAACCTTCACTTATTTGGATTATTTATGTAGAAGTCCAGATCAAAAAG  
CGAAAAGGAATGGCTAGATATTTGCACAAGAAGTAACATAGTCAAATTCAGGAGCA  
CTTAAGCACACATTGAGTGTCTTTTATTTGGAATTTCTTAGATATGGAACTGATGTTTCC  
AAGGGAAGGGTCTATGAGGCAGAGTGAATGTATAGACTGGCATATGGTTAAGT  
GATCATTTGGACTGCCGTTCTACTCCGGTTGTCGTTAACGC

Nucleotide sequence of full-length CPK-2 from *Physcomitrella patens*

(SEQ ID NO: 26)

ATCCCGGGGAACTGCGATCTGAGATTCCAACCTGGAAGGGCCCTCGCGTAAGACCG  
GATCTCGTTTTCTTACGCTTTTGGCCCTCGCGATATTTGTACATTGTTTCCCTCGGTTTT  
ATTCCGATTCGCGCTCTGAAAATGTGAACGGCTGCAAGCTTGGTTTTGGAGCAACGT  
TGGAGCATTAAGGGTTGCGCTCGTCCCTGCCATTCTCGCTTCTGCTCTGGCCTAT  
GTACATGACGACGTAAGGAGAGGATTTGAGGGTTTTGTAAGTGATATAATCCTCCCC  
GAGGAGATTTCTGTGAGTTGATTAACCTGGATCAGCGACATGGGGAAACACTAGTTCG  
AGGGATCGAGGAAGTCCACTCGGCAGGTGAATCAGGGAGTCCGGTCTCAAGACAC  
CCGAGAGAAGAAATGATAGCGTCAATCCAAGACAGACAGGGTGGTAGCGTTGGCG  
CAAACTACTATGGCGAAAGCCAAGCAGTGTGCTCAGGCCGGGAGAACGATCCACC  
TCTGCGCCGCTGCTCTGCGAGGCCAAGCCAGCATCGAGGTGAGTATCCGGTGT  
TTGGGTAAGCCGCTGTGATAGTATTCGTCAATCTTACATCTGGGACGGGAGCTTGGC  
CGAGGGCAGTTCGGAGTGACTTACTTGTGTACTGACAGAATGAGCAATGAGGCGTA  
CGCGTCAAGAGCATCGCCAAACGAAACTGACCAAGGAGGATATCGAGGATG  
TTAAGCGGGAGGTTCAAGATTATGCATCACCTGTGCGGGACACCAATATCGTGGTGT  
TAAAGGATGTGTTGAGGACAAGCATTCCGTGCATCTTGTGATGGAGCTCTGTGCAG  
GTGGCGAGCTCTTCGATCGCATATTGCCAAGGGGCATTACAGTGGAGCGCCCGCTG  
CGATATGTGCAGAGTCAATCGTCAATGTGGTGCACAGATGCCACTCATTAGGGTCT  
TCCATCGGGATCTCAAGCCAGAGAATTTCTGTTGGCCAGCAAGGCTGAGGATGCGC  
CTCTGAAGGCCACAGACTTCGGTCTGTCACTTTCTTTAAGCCAGGAGATGTGTTCC  
AGGATATTGTTGGAAGTGCATATACGTGGCCCTGAAGTTTTGAAGAGAAGTATG  
GTCTGAAAGCTGATGTTTGGAGTGCAGCGCTGATGTGTACATCTGTGTTGGTGTG  
TACCCCTTCTGGGCTGAACTGAGCAGGGTATCTTTGACGCTGTGCTCAAAGGGC  
ACATAGACTTCAGAAACGATCCATGGCCGAAAATCTCCAACGGGGCTAAGGATTTG  
GTGAGGAAAATGCTAAACCTTAACGTGAAGATACGTCTGACGGCACAGCAGGTGTT  
GAACCATCCATGGATGAAGGAAGATGGTGTGCTCCAGACGTGCCACTCGACAATG  
CGGTGTGACCAAGTCAAAAATTTCTCAGCCGCAACAGATGAAAAGCTGGCG  
CTGAAGGTGATTCAGAGAGTCTGTGCGAGGAAAGATCGTGGGGTTGAGGAGAT  
GTTCAATCCATAGATACAGACAACAGCGGCACGGTACGTTCCAGGAGCTTAAGG  
AAGGTTGCTGAAGCAGGGCTCAAACTTAATGAATCGGACATCAGGAAACTAATG  
GAAGCTGCAGATGTGATGAAAACGGCAAGATCGACTTCAACGAGTTTCAATCGGC  
AACATGACATGAACAAGACGGAGAAAGAGGATCACCTTTGGGCGCATTATGTC  
ATTTGACACGGACAATAGCGGGTATATCACCATCGACGAGCTCAGGAACTAATG  
GAGAAGAAATGGAATGGAGATCTGAGACCATCCAAGAGATCATCAGCGAGGTGGA  
CACAGACAACGACGGAAGAATAGACTACGACGAGTTGATGCCATGATGCGCAAGG  
GCAATCTCGCCGCTGAAAACGGAGGAACGGTGAACAAGCCAGACACAGGTAGTA  
GCTCCTGGTTGCCAATTTGACGACGGGTTGGCAAGGCAACAGTAGTTGTTGTTAGC  
TTTCAGATTCAGGTTCCGTTATGTTTATGCCCCCTTTGTCTCGAACAAATGGACTCTA  
GGCTTTCCAATGGAAAAGCTATTCACAACGGGTTGCATAACGTGTAGTAGAATGA  
AAGCATTGCTTGGGGGTGACAGTGCCTGTGATCTTGTGGAGTCTCGTAGGATGG  
CTTCGGTTGGATCTCGTTAACGC

Deduced amino acid sequence of PK-6 from *Physcomitrella patens*

(SEQ ID NO: 27)

MGVDMKAPAKQSLGVGLLLCSVIVLSVSSVYGVQVQTPVDTTGLISMWYDLKQSQSL  
TGWTQNASNPGQWYGVVCDGSSVTEIKIGSRGLNGFNPNPSYFQNAFKLRIFDASN  
NIEGNIPOQFPPTSLTQMI LNNKLTGGLPQFDQLGALTVVNLNNSNNL TGNMNPYFNVI  
NVETFDVSYNQLQEGTLPDS ILNLAKLRFNLQNNKFNKGLPDDFSRLKNLQTFNIENDQF  
TGNYPGLPNSRVGNRLTFPPPPAPGTPAPRTPPSPGTSNGSSSHLPLGAIIGIAAGGAV  
LLLLLALGICLCRCRKS KALGDPEATTSRRPWFPTPLS AKSQSDPSKSIDKTKRNI FGS  
SKSEKKS KHRVFEPAFLDKGADEPVVKA SPPVKVLKAPPSFKGISGLGAGHSKATIGK  
VNKSNI AATPFSVADLQAATNSFSDNLI GEGSMGRVYAEFPNGQVLAVKKIDSSASM  
VQNEDDLFLSVVDSLARLQHANTAEVLGYCI EHDQRLLVYEVYVSRGTLELHLSGENTK  
ALSNNVRIKIALG SARALEYLHEVCAPPVHHNFKSANILLDDLNPHVSDCGLAALAPS  
GSRQVSAQMLGSPFGYSAPYAMSGTYTVKSDVYVFGVVMLELLTGRKSLDSSRRPSEQ  
SLVRWATPQLHDIDALARMVDPSLKGITYPAKLSLRFADIVALCVQPEPEFRPPMSEVVA  
LVRLMQRASLSKRRSESAVGIENEPSETS

Deduced amino acid sequence of PK-7 from *Physcomitrella patens*

(SEQ ID NO: 28)

MSVSGMDNYEKLEKVGEGTYGKVIKARDKRSQQLVALKTRLEMEEEGVPSTALREV  
SLQLMLSHSMYIVRLLCVHEHVEKSKPMLYLVFEYMDTLKKYIDLHGRGSPGKPLPPK

## APPENDIX-continued

VVQSFMYQLCTGLAHCHGHGVMHRDLKPQNLVLDKQTRRLKIADLGLGRAFTVPMKS  
 YTHEIVTLWYRAPEVLLGATHYSLPVDIWSVGCIFAEIVRKMPLFTGDELQQLLHIFRLL  
 GTNETIWPVGSQHRDWEHFPQWRPQDLSLAVPGLSAVGLDLLAKMLVFEPKSRISAKA  
 ALSHTYFADVDKTAT

Deduced amino acid sequence of PK-8 from *Physcomitrella patens*

(SEQ ID NO: 29)

MADAKEELALRTEMHWAVERSDVGLLRITLKKDKQLVNAADYDKRTPHLHIAASLDCVP  
 VAKVLLAEGAELNAKDRWGKSPRGEAESAGYMEMVKLLKDYGAESHAGAPRGHVESL  
 IQVAPPLPSNRDWEIAPSEI ELDTSELIGKGAFGEIRKALWRGTPVAVKTI RPSLSNDRMVI  
 KDPQHEVQLLVKVRHPNIVQFLGAVTRQRPMLLVTEFLAGGDLHQLLRSNPMLAPDRIV  
 KYALDIARGMSYLHNRSKPIIHRDLKPRNIIVDEEHKLVGDFGLSKLIDVKLMHDVYKM  
 TGGTGSYRYMAPEVFEHQPYDKSVDFSGMILYEMFEGVAPFEDKDAYDAATLVARD  
 DKRPEMRAQTYPPQMKALIEDCWSPYTPKRPPFVEIVKLEVMYEDCLLRLPKDRRHLR  
 DILHLRRNPADS

Deduced amino acid sequence of PK-9 from *Physcomitrella patens*

(SEQ ID NO: 30)

MKRYQRKVKQLRGREGQVLLERTLFFKQLRPSFPVPHLLATPIDSDNVALVLCVLAGPL  
 ELLLRSPLDENSARFLVANVVLAVELLHKDGVVYRGISPDVLMIDRKGRLQLVDFRFAK  
 QMSDERFTTVCGMADFLAPEIQGQGHGLASDWWAVGVLVYFMLQTELPFGSWRDNE  
 LEIFGRITARRQLTFPSSFSPEAVDLIDKLLVVDPTKRLGCDSHGSLAIREHPWFRGINWDK  
 HLDCSVEVPSEIMTRLQLAIDFLPVDDSYQVFDLQPPDEDDPPWLDGW

Deduced amino acid sequence of CK-1 from *Physcomitrella patens*

(SEQ ID NO: 31)

MDLGGDRMRAPQRSREYQYRSLDVFTQHEQLKQKQQQDEYQRTLEKLETLPKMLS  
 NATVSSSPRSPDGRRRLRTVANKYAVEGMVGSAGFCVKYQGSDLTNHEVVGIKLEDTR  
 TEHAQLMHESRLYNIIRGGKGVNMRWFGKEQDYNVMVLDLGGPNLLHLFKVCGLRFS  
 LKTVIMLGYQMDRVEYVHSRGLVHRDLKPDNFMGCGRQGNQVFIIDFGLAKEYMDP  
 ATRRHIPYRDRKSFDTGTAR YASRNQHRGIEHSRRDDIESLGYILMYFLRGNLPWQKGG  
 QRLTDQKQHEVMHNKIKMNTTVEELCDGYPSPQFADFLHARSGLGFYEQPDYCYLRSLF  
 RDLFIQKKFQLDHVYDWTVTYQLPQNGSLQSVRSQNSAASHLQNRPSNVSYCPPLTKS  
 EFRREVVAAN

Deduced amino acid sequence of CK-2 from *Physcomitrella patens*

(SEQ ID NO: 32)

MEPRVGNKYRLGRKIGSGSFGIEIYLGTVNQTNNEEVGKLESIKTKHPQLLYESKLYRILQG  
 GTGIPNIRWFGIEGDYVNLVLDLGLPSLEDLNFPCSRKFSLKTVMMLADQLINRVEYVHA  
 KSPFLHRDIKPDNFMGLGRRANQVYIIDFGLAKKYRDPSTHQHPIYRENKNTGTARYAS  
 INTHLGIEQSRDDLESGLYVLMYFLRGLSPWQGLKAGTKKQYKYEKISEKKMSTPIEVLK  
 KNYPSEFASYFHYCRSLRFDDKPDYAYLKRIFRDLFIREGFQDFYVFDWTILKYQQSQISG  
 GSSTRLGASAGQTSALGTGATGSRDLQRPTEPMDPSRRRLPGGANGSGVANALDSSKH  
 KSPGLDESAKDSALAVSEPERMHTSSYATRGGSSSRRAVLSRRPSSGASAEVVDSSRTG  
 SSKLGPSTLRSAGMQRSSPVTSDPKRISRRHPQPPSANLRIYEAAIKGVESLSVEVDQSR  
 YK

Deduced amino acid sequence of CK-3 from *Physcomitrella patens*

(SEQ ID NO: 33)

MSKARVYTDVNVQRPKDYWDYEAALTQWGDQDDYEVVRKVGGRKYSEVFEVGNVAV  
 NSERCVMKILKPKVKKKIKREIKILQNLCGGPNIVKLLDVRDQSQKTPSLIFEYVNNTFD  
 KVLVYPTLTDQFDIRYIHELLKALDYCHSQGIMHRDVKPHNVMI DHEQRKRLRIDWGLAE  
 FYHPGKEYNVRVASRYFKGPELLVDLQDYDYSLDMWSLGCMFAGMIFRKEPFYGHND  
 YDQLVKIAKVLGTDELMSYLNKYRLELDPHLEALVGRHSRKPWSKFINADNQLRVVPEA  
 VDFLDKLLRYDHQDRLTAKEAMAHYPYFYPVKVSEVSNRRA

Deduced amino acid sequence of MPK-2 from *Physcomitrella patens*

(SEQ ID NO: 34)

METSSGTPELKVISTPTYGGHYVYVAVAGTDFEVTARYKPLRPIGRGAYGIVCSLFDTV  
 TGEVAVKIKGNAPDNIDAKRTLREIKLLRHMHDHENVAITDIIRPPTRENFNVDYIYVE  
 LMDTDLHQIIRSNQALTEDHCQYFLYQILRGLKYIHSANVLHRDLKPTNLLVNCNDLKI  
 ADFGLARTLSETDFMTEYVTRWYRAPELLNCSAYTAAIDIWSVGCIFMELLNRSALFP  
 GRDYVHQLRLITELIGTPEDRDLGFLRSDNARYIKHLPRQSPIPLTQKFRGINRSALDLVE  
 KMLVDFPAKRITVEAALAHPYLASLHDINDEPASVSPFDFEFPPISEEHKDLIWRREALD  
 CSLGPDMMVQ

Deduced amino acid sequence of MPK-3 from *Physcomitrella patens*

(SEQ ID NO: 35)

MGLTPFSCVTVQGYVRYVYPDGHVENLSKSCSVHDLGLGNPDYVYCGSTPYTITNRMA  
 AEEVLEYGVTVFCATPNAQPFLEERQPKVHRGSKILPRFSKHGVHRELSPTHGSQQS  
 RKVFDYHSVTMQQLESIRNEGPEPHLAGDRPSKHLKLVFIRHCLRALRLPRISIDLMSPL

## APPENDIX-continued

---

PNLSGEALSPTATAKDEITQMILKSAABSELGMYVSKRQEFYLRARRRRRFAWKPVLQ  
SISEMKPVMEFHTPMAYRDSGSPKKNASTPSLPGPKNISPPRQVSVQRSSPPKKNVSPFP  
QPAFVARTASKYSAASQVQRNRGNKSLYMA

---

Deduced amino acid sequence of MPK-4 from *Physcomitrella patens*

---

(SEQ ID NO: 36)

MSRRVRRGGLRVAVPKQETPVSKFLTASGTFQDDDIKLNHTGLRVVSSEPNLPTQTQSSS  
PDGQLSIADLELVRFLGKAGGTVQLVRHKWTVNRYALKAIQMNINETVRKQIVQELKI  
NQVTHQQCPYIVECFHSFYHNGVISMILEYMDRGSLSDIIKQKQIPEPYLAVIASQVLKG  
LEYLHQVRIIHRDIKPSNLLINHKGEVKISDFGVSAVLVHSLAQDRTFVGTCTYMSPERL  
QGRSYAYSDLDLWGLTLLECALGTFPPYKPMGMEEGWQNFILMECIVNQPPAAASPK  
FSPEFCFSIESCIRKCPSERPSTDDLKHPFLQKYNEEYHLSKIL

Deduced amino acid sequence of MPK-5 from *Physcomitrella patens*

---

(SEQ ID NO: 37)

MSRRVRRGGLRVAVPKQETPVSKFLTASGTFQDDDIKLNHTGLRVVSSEPNLPTQTQSSS  
PDGQLSIADLELVRFLGKAGGTVQLVRHKWTVNRYALKAIQMNINETVRKQIVQELKI  
NQVTHQQCPYIVECFHSFYHNGVISMILEYMDRGSLSDIIKQKQIPEPYLAVIASQVLKG  
LEYLHQVRIIHRDIKPSNLLINHKGEVKISDFGVSAVLVHSLAQDRTFVGTCTYMSPERL  
QGRSYAYSDLDLWGLTLLECALGTFPPYKPMGMEEGWQNFILMECIVNQPPAAASPK  
FSPEFCFSIESCIRKCPSERPSTDDLKHPFLQKYNEEYHLSKIL

Deduced amino acid sequence of CPK-1 from *Physcomitrella patens*

---

(SEQ ID NO: 38)

MGCYCGKFDGEGEDLFRQKQVSRTPKHGSWSNSNRGSFNNGGASPMRAKTSF  
GSSHPSRPHSASPLPHYTSSPAPSTPRRNIFKRFPFPPSPAKHIQSSLVKVRHGAKPKEGGAI  
PEAVDGEKPLDKHFGYHKNFATKYELGHEVGRGHFGHTCYAKVRKGEHKGQAVAVKII  
SKAKMTAIAIEDVGREVKILKALTGHQNLVRFYDSCEDHLNVIYVMELECEGELLDRIL  
SRGGKYSEEDAKVVRQILSVVAFCHLQGVVHRDLKPENFLFTTKDEYAQLKAIDFGLS  
DFIKPDERLNDIVGSAYYVAPEVLHRLYSMEADVWSIGVIYIILLGSRPFWARTESGIFR  
AVLRADPSFEEAPWPSISPEAKDFVKRLLNKMRRMTAAQALTPWIRSNVVKIPLDIL  
VYRLVRYLRASMRKAALKALSKTLTETEFTYLRQFMLEPSSNNGRVTFENFRQALL  
KNSTEAMKESRVFEILESMDGLHFKKMDSEFCAAAISVLQLEATERWEQHARAAYDIF  
EKEGNRVIIYPDELAKEMGLAPNVAQVFLDWIRQSDGRLSFTGFTKLLHGISRAIKNLQ  
Q

Deduced amino acid sequence of CPK-2 from *Physcomitrella patens*

---

(SEQ ID NO: 39)

MGNLSSRGRKSTRQVNQGVGSQDTREKNDVSNPKTRQGGVSGANNYGGKPSGQA  
GERSTSAPAALPRPKPASRSVSGVLGKPLSDIRQSYILGRELGRGQFGVYTLCTDKMTNE  
AYACKSIKAKRLTSSKEDIEDVKREVQIMHLSGTPNIVVLKDVFPEDKHSVHLVMELEAG  
GELFDRIIAKGHYSERAAADMCRVIVNVVHRCHSLGVFHRDLKPENFLASKAEDAPLK  
ATDFGLSTFPFKPGDVPQINVGSAYYVAPEVLKRSYGPVADVWSAGVIVYILLCGVPPWF  
AETEQGIFDAVLKGHIDFENDPWPKISNGAKDLVRKMLNPNVVKIRLTAQQVNLNHPWMK  
EDGDAPDVPDLNVAVTLRKNFSAANKMKLALKVIAESLSEEEIVGLREMPKSIDTDNSG  
TVTFEELKEGLLKQGSKLNESDIRKLEAADVNGKIDFNEFISATMHMNKTEKEDHL  
WAAFHFDTDNSGYITIDELQEAMEKNGMDPETIQEIISEVDTDNDGRIDYDEFVAMM  
RKGNPGAENGGTVNKRHR

---

## SEQUENCE LISTING

<160> NUMBER OF SEQ ID NOS: 128

<210> SEQ ID NO 1

<211> LENGTH: 695

<212> TYPE: DNA

<213> ORGANISM: *Physcomitrella patens*

<220> FEATURE:

<221> NAME/KEY: modified\_base

<222> LOCATION: (636)

<223> OTHER INFORMATION: a, t, c, g, other or unknown

<220> FEATURE:

<221> NAME/KEY: modified\_base

<222> LOCATION: (680)

<223> OTHER INFORMATION: a, t, c, g, other or unknown

<400> SEQUENCE: 1

gcacgagctc aatcctcatg tttcggactg tggactagct gcccttgca ccatctggttc 60

-continued

---

```

tgaacgccag gtgtcggcac aaatggtggg ctctttcggg tacagtgcc ctgagtacgc 120
catgtctgga acctataccg tgaagagtga cgtctacagc ttcgggtgtg taatgctgga 180
gctactcact gggcgcaagc cttagacag ctcaagacca cgatccgagc aatctttggt 240
acgatgggccc acacctcaat tgcacgacat cgacgccctt gcacgaatgg tggatccgtc 300
ggtgaagggc atctaccctg ctaaatcact ctctcggttt gctgatatag tegccctttg 360
cgtccagccc gagccccagt tccgaccccc gatgtctgaa gtggtgcagg cacttgtaag 420
gctgatgcag cgtgcgagtc tgagcaaacg cagatcggag tccgctgttg ggaattgagt 480
cgaacgagcc atctgagact tcacctttga gactactgaa gcgccacta gctaatacgt 540
gcatctttgg ccatctcgtt tctgagtgga acacaaagct gggatatattc tttggtggtt 600
aagcaaccat ttgtcccaat ttgaacttcc gctggngaag gtctgtatgt tgagaaacga 660
tgcaaacgct tcgctggtn tgcttgaact tcaaa 695

```

```

<210> SEQ ID NO 2
<211> LENGTH: 512
<212> TYPE: DNA
<213> ORGANISM: Physcomitrella patens

```

```

<400> SEQUENCE: 2

```

```

ggcacgagcc gaacttcagc agcttcttca catcttcagg ttgcttgga ccccgaaatga 60
gacaatctgg cctggtgta gccagcaccg tgattggcac gagtttctc aatggagacc 120
acaagatctg tccttctgct tccccgact cagcgcggtt ggcttagacc ttctcgccaa 180
aatggtggta ttcgagccct caaagagaat ctctgccaaa gccgcctga gccatactta 240
tttcgctgat gttgataaga cagcaaccta aacacaacag aacaattcaa gagaaccagg 300
taacctctac ctgtccaaga cgaaggacat ctaactcttc agtcaaactt ggccaatcat 360
gctgattggg aattgaacca caggaacgag gtgggcaccg tggttcgctg tagcatacaa 420
agtagtctgg aagacttgac atcgttagct ggcaatgcag tattttgaa atacaatttt 480
tcattaaaaa tctcctaaag attcaatatt tg 512

```

```

<210> SEQ ID NO 3
<211> LENGTH: 651
<212> TYPE: DNA
<213> ORGANISM: Physcomitrella patens

```

```

<220> FEATURE:
<221> NAME/KEY: modified_base
<222> LOCATION: (608)
<223> OTHER INFORMATION: a, t, c, g, other or unknown

```

```

<400> SEQUENCE: 3

```

```

gcaccagact atgacaagcg cacgccttg cacatcgcg cgtccctgga ttgtgtccct 60
gttgctaaag tcctgcttgc ggaaggagca gagttgaatg caaaagacag gtgggggaaa 120
tctccgagag gcgagggcga gagtgcagga tacatggaga tggtaaagct gttgaaggat 180
tacggggctg agtcacacgc aggtgccccg aggggccaccg ttgagagtct gattcaggtt 240
gccccctcgt tgcttctaa ccgcgactgg gagatcgtc cgtcggagat tgaacttgat 300
accagcgagc tcacgcgcaa aggctccttt ggagagattc ggaaggcgtc ttggcgcgcc 360
acacccgtcg ctgtgaagac aatcagacct tctctgtcca acgacagaat ggtcatcaag 420
gacttcagc acgaggtgca attgctcgta aaggttcggc acccaaacat tgtgcagttc 480
ctcggggctg ttaccctgca aagacctctc atgttagtca ccgagtttct ggcagggggg 540
cgatttgcat cagttgctga ggagcacct aaatttgct cctgaccgca tcgtgaagta 600

```



-continued

---

 tgcctcnac atagctcgcg gcatgtctta cttcaccatc ggagcagccc a 651

<210> SEQ ID NO 4  
 <211> LENGTH: 710  
 <212> TYPE: DNA  
 <213> ORGANISM: Physcomitrella patens  
 <220> FEATURE:  
 <221> NAME/KEY: modified\_base  
 <222> LOCATION: (54)  
 <223> OTHER INFORMATION: a, t, c, g, other or unknown

&lt;400&gt; SEQUENCE: 4

tccagcccat ttggttgccc acacacagct gttcatgagt caccgcttc aggntgaact 60  
 gaagaaacgt aactccgtac ggctatttta ccaaattttc aagctcgttg tcccgccatg 120  
 atccaaatgg aagctcagtt tgcaacatga agtacattga acacacctac cgcccaccag 180  
 tcagaagcca ggccatgacc ttgtccttga atgatctcgg gtgctaagaa atcagccatg 240  
 ccacagactg taaaagtgcg ctcacccgac atttgctttg caaacggaaa atcaaccaggc 300  
 tgaagtcgct ctttccgac taccataaga acatcgggag agatgccacg atatacaacg 360  
 ccatccttgt gcagaagttc gacggctaata accacgcttg cgaccagaaa acgagctgag 420  
 ttctcgtcta aaggtgaccg aagtagaagt tctagaggcc cagctaacac acaattaaga 480  
 acgagtgcca cattgtcact gtcaaatagg gtggccaaga gatcggcac gaatggggaa 540  
 ggctcagtt gcttgaaga agttctctcc aataggactt ggccctccc accgagctc 600  
 tgaactttac gtctctgcta cttttcatg cttatgacgt catctgattt cttgcagagc 660  
 accacaccga catcacagca atcggttgaa tagacctggt gccgattcct 710

<210> SEQ ID NO 5  
 <211> LENGTH: 1271  
 <212> TYPE: DNA  
 <213> ORGANISM: Physcomitrella patens  
 <220> FEATURE:  
 <221> NAME/KEY: modified\_base  
 <222> LOCATION: (619)  
 <223> OTHER INFORMATION: a, t, c, g, other or unknown

&lt;400&gt; SEQUENCE: 5

tatgcccatc ttctcact cagaccagat cctctatttc aattacagaa gaaagttgct 60  
 tgtgcaacgt attgaaatca tcaccgtcat gggctttccg agtaaaaatt cttgtaatgg 120  
 ataaagtcat ttctagtctg atccatacaa gctaccgaca caatgctaga agccttgatt 180  
 tacacactac acactagaga gtctacaact cttttctac actctgctta gttgcctcat 240  
 cctcaactcc ataaaccccc attcacaatc atgtaagact tgagagaggg aaacagtaag 300  
 caaccttgty ctattttagt accagagcag aggatgaacc actagtcctc ccaacgtaag 360  
 ccctaattcg ccgcaacaac ctcacgacgg aactccgact tggtaaggg tggacaatat 420  
 gatacattcg aaggtcgatt ttgcaaatgg gacgaagcag cggattctg gctgcccact 480  
 gattgcagag agccattctg ggggagttga gtatacacag tccagtcgta cacatggctg 540  
 agctggaatt ttttctgaat gaaaagatca cggacaagc ttcggaggtg cagtagtcag 600  
 gctgctcgta aaaacctana cttcggcgcg tgggcaaaa agtcggcaaa ttgactggga 660  
 taccatcac aaagctcctc ccacagtggg ggtcatcttg atttgtgtg gcatgtactc 720  
 gtgttgcttc tggctcagtg gggcgttgcc cgcccttccc ttgccatggc aaattgctc 780  
 ttagaaagta cataagaatg taaccaagt gattctatgt catctctct actgtgctcg 840  
 attcctctgt gctgattcct actagcgtac cgtgcccctc ctgtgaagct cttcctatct 900

-continued

---

cggttaagga tatgccttcg tgttgccggg tccatgtact cctttgcaa gccaaaatct	960
ataatgaaca ctgtgtttcc ttgccgaccg cagcccatga ggaagttatc cggcttcagg	1020
tcacggtgaa cgagccctcg agaatgcacg tattccaccc ggtcaatcat ttgtaaccg	1080
agcataatca cggctcttcaa cgaaaacctt agcccacaca ccttaaagag gtgcaacagg	1140
ttcggcccca ataggcttag caccatcaca ttgtagtett ctgctgcttt tccgaacct	1200
ctcatgttg gcaactcctt cccaccccg aatatgttg acaagcgga ctgctgcatt	1260
aactctcgtg c	1271

&lt;210&gt; SEQ ID NO 6

&lt;211&gt; LENGTH: 1910

&lt;212&gt; TYPE: DNA

&lt;213&gt; ORGANISM: Physcomitrella patens

&lt;400&gt; SEQUENCE: 6

ttttttttt ccaatagatt tgcattacat aactccaagt tatgatatgt acaggtttagc	60
aacaagctaa tggctgcaag cagtgaacat actaccaagg gagagattct cactccctag	120
acttcatcct cgtacgttac ttggcaagga ttatggttta gtgataaaaa gcttcacaag	180
ccggcaagca tgcgtgttgc ttctgtgca atctaataatg tatttcctta ggaatcgtat	240
ggcagagagc taccacacaa agcactgaca atggtttgat ggtaacaaga tagagatcca	300
ttcattccta agtatgagag acctgtatgc ttagcaccat tgtaggacag aaccaccgtt	360
ttcccctcaa tcaggctgtt gccaaatgta gagcaactct catcaacata acaagagggt	420
ttgatagaag acagagcccc gctatataac cacaagcctt gcgcctacct tataacggct	480
tggatccacc tcaacagaaa gtgattcaac toccttgata cgggctttcg taaatcctca	540
agttggcaga tggcggttgt ggatggcggc tagatatccg ctttgggtcc gaagtaactg	600
gagagctcct ctgcacccct gctgacgacc gtaagctggt gggaccaagc ttactgctcc	660
ctgttcgaga ggaatctacg acttctgctg atgccccga gggcctgctg ctagatagga	720
cagctcgcct ggagaagaa ccccccgag ttgcatacga agatgtatgc atgcgctctg	780
gttctgacac aacagcaaga gcagaatcct tagcagatc atcaagtcca ggacttttgt	840
gcttagatga gtccaaaagca tttgcgacc cggagccatt tgcctctcca ggaagcctgc	900
gccgagaagg atccattggt tcggtgggcc gctgcagtc tccgcttctc gtagccccag	960
ttccaagtgc accactggtt tgccctgcag aagcaccag tgcagttgaa ctgccaccg	1020
aaatttgtga ctgctggtac ttcagaatg tccagtcaa aacgtagtca aattgaaaac	1080
ctgtaaaact atttccagt taggcaaaca gaagtggcac tgtaataaac tgaaaatcat	1140
caaacattca caaactatct gttcgttgat agagcatagt aaagtctgcg cttaggatca	1200
agtcttgata cattacaatg cccaagcaag agtgaaacct acaaaagtta cagttttcat	1260
accctcacga ataaagagg cacggaagat tcttttcaa tatgcatagt cgggtttgtc	1320
atcaaaaagc aaggaccggc agtagtgaa gtacgctcgt gcgaattctg aaggataatt	1380
tttcaaaag acctcaatgg gcgtggacat ttgtttctc actgatcttc tctacttct	1440
gcttcttgg tcccgtttc agtccctgac catggaagac tgctctcag gaagtacatg	1500
agcacatata caagagatc caaatcatct cgtctgcttt gctcaatacc aagatgagtg	1560
ttgatgctg cataccgagc agtccctgac agatttttgt tctccctgta gggaaatagc	1620
tgatgcgtgg aagggtcgcg gtacttctg gcaagaccaa aatcaataat gtagacctgg	1680
ttgctcgc taccagccc cattagaaaa ttatcaggct tgatgtctct atgaagaag	1740

-continued

---

```

cttttcgcat gcacatactc cactctgttg atcagctggt cagcaagcat gagaacagtc 1800
tttaaagaga acttccggct gcagaagttg aaaaggtctt cgagacttgg ccccaacaga 1860
tccagaacca agacattgta gtctccttct atccccgaacc atcctcgtgc 1910

```

```

<210> SEQ ID NO 7
<211> LENGTH: 720
<212> TYPE: DNA
<213> ORGANISM: Physcomitrella patens
<220> FEATURE:
<221> NAME/KEY: modified_base
<222> LOCATION: (58)
<223> OTHER INFORMATION: a, t, c, g, other or unknown
<220> FEATURE:
<221> NAME/KEY: modified_base
<222> LOCATION: (613)
<223> OTHER INFORMATION: a, t, c, g, other or unknown

```

```

<400> SEQUENCE: 7
cggtggggcg ctccccaaata ttttatcccc ggggctgcag ggaatccggc gaccagtntt 60
tgaaggtgtc aacgcctgta atagtgcgcy ttgcgttatg aagattttga agccagtaaa 120
gaaaaaaaaag atcaaaagag agatcaagat tctgcaaaac ctttgtggag ggcccaacat 180
tgtgaagctt ctggacattg tccgtgatca gcaatcgaag acaccagcc taatttttga 240
gtatgtgaac aatactgatt tcaaagtgc ctaccccact cttacagact ttgatatccg 300
atactacatt catgagctgc tcaaggcttt ggactattgc cattctcaag ggattatgca 360
cagggatgtg aagccacaca acgtgatgat tgaccatgag cagcggaaagc ttaggcttat 420
tgactgggga cttgcccgaat tctatcatcc tggcaaagag tataatgtgc gtgttgccctc 480
taggtacttc aagggtcctg agctgctggt tgatcttcaa gattatgatt actctctcga 540
catgtggagc tctgggggtgc atgtttgccg gcatgatatt tccgaaggag ccattctttt 600
atgggcata canttcatga tcaacttggg gaagatcgct aagggtgttg gaacttgatg 660
aattgaatc ctatctaaca aataccgcta agtggacccc attggagcac ctggtggggg 720

```

```

<210> SEQ ID NO 8
<211> LENGTH: 953
<212> TYPE: DNA
<213> ORGANISM: Physcomitrella patens

```

```

<400> SEQUENCE: 8
gcacgaggaa ctaacgaatt gtcattctat aatccaatag tgtaatcaca cgggggggaa 60
taagttgcaa aaccatacaa cgccgggata gcggtgtagc cacctaaaga attgagagta 120
ggccttacia cttgagatga agtgtgaagt ggtactgcac catatcatca ggacctaaagc 180
tgcaatccag agcctccctc caaatgagat ccctgatagg ctctccgag atagagggct 240
cctcgaagcc aaactcgaag ggagataccg agccaggctc atcgttgatg tcatgaagtg 300
aagcttaaat aagggtgcgc caagcagct tccactgtga ttcttttcgc tggatcaaag 360
accgcatct tttcaacaag atcaagagca gaacgattaa tgctctgaa cttctgggtt 420
aagggaatag gcgactgtcg aggcagggtc ttgatatacc gcctagcatt gtcgcttctc 480
aaaaacccaa gatccctatc ttcaggagtt ccgatgagtt ctgtaattag gcggagctga 540
tgacatagtc ctctcccagg gaacaacgca gatcgggttaa gcaactccat gaagatgcac 600
cccacagacc aaatgtcaat agctgcagtg tatgctgaac aattcaggag cagctctgga 660
gctctgtacc acctcgttac aacatactca gtcataaat ccgtttcaga gagagtgcgt 720
gccaagccaa aatctcggat tttcaaatcg caattggcat tgacgagaag gttggtgggc 780

```

-continued

---

```

ttcaagtccc ggtgcaagac gttcggcga tggatgtact tcaagccccg caagatttga 840
tacagaaaat actgacagtg gtcttctgtg agagcttgat ttgaacgaat gatctggtgt 900
aggctccgtat ccatcaactc gtatacaatg tacacgtcgt tgaatctctg tgc 953

```

```

<210> SEQ ID NO 9
<211> LENGTH: 683
<212> TYPE: DNA
<213> ORGANISM: Physcomitrella patens
<220> FEATURE:
<221> NAME/KEY: modified_base
<222> LOCATION: (663)
<223> OTHER INFORMATION: a, t, c, g, other or unknown
<220> FEATURE:
<221> NAME/KEY: modified_base
<222> LOCATION: (670)
<223> OTHER INFORMATION: a, t, c, g, other or unknown

```

```

<400> SEQUENCE: 9
cggcaccagc ctgctgggag accgaccatc gaagcacctt aagctcgttt tcattcggca 60
ttgcttgcca gcacttcgac ttcctagaat ttcaatagac ctaatggaat cgccactccc 120
taatctttcc ggagaggcct tatcgccgac ggcaactgcc gaagacgaga ttactcagat 180
gatactaaaa agtgccgcaa ggtccgaatt aggaatgtat gtttcgaaga gacaggaatt 240
ctatcttcga agagcgcgga ggcggcgtaa gtttgctggg aagccggttt tgcagagcat 300
ctccgagatg aagcctgtca tgggaattcca cactccgatg gcttaccggg atagtgggtc 360
tccgcccgaag aagcctccta ccccatcctt acctggcccg aagaacattt caccgccacg 420
acaagtgagt gtcccgcaaa ggagcagtc tccgcccgaag aacgtctcac cacctcccca 480
gcccggcatt ttgtagcgcg gactgcgatc gaagtattct gctgcatctc agcaagtcca 540
acgaaatcga gggcaacgcg aaatctcttt tatatggcgt agtttgtgtc tccgactgga 600
ctctatccta tccccatcga gataactgat tcggtggata atttctccaa attttggtc 660
acncaagaan ctcaagggcg aat 683

```

```

<210> SEQ ID NO 10
<211> LENGTH: 1156
<212> TYPE: DNA
<213> ORGANISM: Physcomitrella patens
<220> FEATURE:
<221> NAME/KEY: modified_base
<222> LOCATION: (923)
<223> OTHER INFORMATION: a, t, c, g, other or unknown
<220> FEATURE:
<221> NAME/KEY: modified_base
<222> LOCATION: (1143)
<223> OTHER INFORMATION: a, t, c, g, other or unknown

```

```

<400> SEQUENCE: 10
gcacgaggtt ggtgtaagtt attgatagtg ctgtgcaatt cacagttttg ctactccggt 60
aggtccgacc tcttcaattg tcagtttaaa aactctaaaa acatttgaga aaagtgttga 120
aaaaatctccg tgaggaaatt ccttgtcgca agacgtgaaa aaaagaagaa agaagatgga 180
aatattgttt tgggtatcga agaagtgttc gatgctgtgc aataaggaaa gaaaaagtgc 240
aggtaacata aaaagctagc atggtgatga taatataaga ccccattaa cacacttatg 300
gattgtttca tgagctgcac gttctcagcg acaaatgggg ctattgaga aaactccact 360
ttctataagg ttgggaaacg agcgtttttt ttttgaagat gttttttccg tcaatctgat 420
ttgatatcgt tctcaacttg accacatagc actatataag gaaaaggcat tgagaaagtg 480
gcggattggc gaggtagttc gaccatgctt ttggtaaagt cccttgaagt tcagtgggtg 540

```

-continued

---

atcaggcttg	tggtagtgc	agtctctgca	cgccatgcga	ggctaacttt	aagttacaaa	600
atcttgctca	aatggctactc	ttcctcgttg	tacttttgca	ggaacggatg	tttaagtaaa	660
tcagtagttg	atggctcgttc	actgggacat	ttccggatgc	aggattcaat	aaaagaacaa	720
aattcggggg	agaatttgc	aggggatgcg	gctgcggggg	gttgattaac	tatacattcc	780
atgaggatga	agaaatttg	ccaacccctc	tccattccag	ctggtttgta	tggaaggta	840
cccaacgcac	actccaaaag	agtcaatcct	aaactccata	ggcactgtc	gtagcatac	900
gaacgccctc	gaaggcgttc	tgncgacata	tatgtgcaag	tcccaacgaa	cgtgtctcgc	960
tgggccaagg	aatgaaccaa	cacagcactg	acacccaaat	cagatatttt	gacctaccc	1020
ttgtgattga	tgaggaggtt	ggagggtttt	atatcacgat	gtagtagtg	cctgacttgg	1080
tgtaggtatt	ccaatccctt	cagaacttga	ctagcaatga	cggccaaaata	cggtcaggt	1140
atntgctttc	tggtgc					1156

&lt;210&gt; SEQ ID NO 11

&lt;211&gt; LENGTH: 629

&lt;212&gt; TYPE: DNA

&lt;213&gt; ORGANISM: Physcomitrella patens

&lt;400&gt; SEQUENCE: 11

tccccgggct	gaggaattcg	gcacgagcgg	ttgatcctca	cccttgggaa	ggaccctgga	60
attgagtage	gtgcggaagc	tgcatcgatc	cggaagagac	gatgagtagg	agagtgagaa	120
ggggaggtct	tcgctcgcg	gtgccgaagc	aagagactcc	cgtcagcaaa	ttttgactg	180
ccagtggaac	tttccaggat	gatgatata	agtcacaaca	caccgggctt	cgcgtcgtct	240
cttcagaacc	taaccttctc	acgcagacgc	agtctagctc	cccagatggg	caactgtcaa	300
tagcagacct	ggagttagtg	cggttcttag	gaaaggggtc	gggtggaacc	ggtgcagctt	360
ggtccggcac	aaatggacca	atgtcaatta	tgcaactgaa	gcgatacaaa	tgaatatcaa	420
cgaaacagtg	aggaagcaga	ttgttcagga	gctgaaaatc	aaccaagtga	cgcaccagca	480
gtgcccttat	atcgtggaat	gcttccactc	cttctaccac	aacggcgtca	tatccatgat	540
cctagagtac	atggacaggg	gctcgttgtc	cgacattatt	aagcaacaaa	agcagatacc	600
tgagccgtat	ttggccgtca	ttgctagtc				629

&lt;210&gt; SEQ ID NO 12

&lt;211&gt; LENGTH: 514

&lt;212&gt; TYPE: DNA

&lt;213&gt; ORGANISM: Physcomitrella patens

&lt;400&gt; SEQUENCE: 12

gcaccagccg	agtcgggcat	ttttcgtcgc	gtgttgaggg	ctgaccagag	ctttgaagaa	60
gccccttggc	cttccatctc	tcccgaagcc	aaggatttcg	tgaagcgtct	cctgaataag	120
gatatgcgga	aacgcatgac	tgctgcacaa	gctttaactc	atccatggat	tcgaagtaac	180
aacgtgaaga	tacctctgga	tatcttagtg	tacagacttg	tgaggaatta	tcttcgtgca	240
tcatccatga	gaaaggctgc	tttgaaggcc	ctgtcaaaga	ctttaaccga	agaacgagact	300
ttttatctac	gtactcaatt	tatgtcgtca	gaaccaagta	acaacggctc	tgtaactttt	360
gagaatttca	gacagccact	gctgaaaaat	tcaacagagg	ccatgaaaga	gtcacggggt	420
tttgaaatte	tggaatcgat	ggatggctct	catttcaaga	aaatggactt	ttcagagttc	480
tgtgcagcgg	ccattagtgt	tctccagtta	gaag			514

&lt;210&gt; SEQ ID NO 13

-continued

---

```

<211> LENGTH: 1387
<212> TYPE: DNA
<213> ORGANISM: Physcomitrella patens
<220> FEATURE:
<221> NAME/KEY: modified_base
<222> LOCATION: (1385)
<223> OTHER INFORMATION: a, t, c, g, other or unknown

<400> SEQUENCE: 13
gcacgagctc ctgcctctcc cctctctctc cctctctc atctctggagc ccagcgaact    60
gcgatctgag attccaactt ggaagggcct cgcgtaagca ccggagctcg tttcttacgc    120
ttttgcgcct cgcgatattt gtacattggt tcctctgggt ttattcgatt ccgcctctga    180
aaatgtgaac gggctgcaag cttggttttg gagcaacggt ggagcattga aggggtgctgc    240
tcgtccctgc ccattctctc cttctgctct ggcctatgtc atgacgacgt gaaggagagg    300
atctgagggg tttgcaagt atataatcct ccccgaggag atttctgtga gttgattaac    360
ttggatcagc gacatgggga acaactagttc gaggggatcg aggaagtcca ctccggcagg    420
gaatcagggg gtcgggtctc aagacacccg agagaagaat gatagcgtca atccaaagac    480
gagacagggg ggttagcgtt ggcgcaacaa ctatggcgga aagcacaagc agtgggtgctc    540
aggccggaga acgatccacc tctgcgcccg ctgctctgcc gaggccgaag ccagcctcga    600
ggtcagtatc cgggtgtttg ggtaagccgc tgtcagatat tcgtcaatct tacatcctgg    660
gacgggagct tggccgaggg cagttcggag tgacttactt gtgtactgac aagatgacga    720
atgaggcgta cgcgtgcaag agcatcgcca aacggaaact gaccagtaag gaggatatcg    780
aggatgttaa gcgggagggt cagattatgc atcacctgtc ggggacaccc aatatcgtgg    840
tgttaaagga tgtgttcgag gacaagcatt ccgtgcatct tgtgatggag ctctgtgcag    900
gtggcgagct cttcgatcgc atcattgcca aggggcatta cagtgagcgc gccgctgccg    960
atatgtgcag agtcctcctc aatgtgggtc acagatgcca ctcatagggt gtcttccatc   1020
gggatctcaa gccagagaat tttctgttgg ccagcaaggc tgaggatgcg cctctgaagg   1080
ccacagactt cggctctgca actttcttta agccaggaga tgtgttcag gatattgttg   1140
gaagtgcgta ttacgtggcc cctgaagttt tgaagagaag ttatggctct gagctgatgt   1200
ttggagtgca ggcgtgattg tgtacattct gctgtgtggt gtacccccct tctgggctga   1260
aactgagcag ggtatctttg acgctgtgct caaagggcac atagactctg agaacgagtc   1320
catggccgaa aatctccaac ggggctaagg atttgggtgag gaaaatgcta aaccctaacg   1380
tgaanat                                           1387

```

```

<210> SEQ ID NO 14
<211> LENGTH: 2784
<212> TYPE: DNA
<213> ORGANISM: Physcomitrella patens

<400> SEQUENCE: 14
atcccggggt agtatcactt acgggtggcga gggatggcct ttggggtagg agctggtata    60
tgcggagtc c aacagaagct tgtgcaggac tcttgagttg tgcgtgagag ggctgagtg    120
cgaaaggtt ttttccgacg aagagtcaat gtgggcgtgg acaaacgttt gaagagatgg    180
gtgtggatat gaaggtccg gctaagcagt cgctgggagt cggactgctc ctgtgctctg    240
tagtgatcct ctccgtgggt agctctgtgt atggccaagt tcagacagat ccagtgata    300
ctacaggctt aatttccatg tggatgact taaaacagag tcaatctctc acggggtgga    360
ctcaaaatgc ttctaaccct tgtgggcagc agtggtaagg cgttgatgt gatggctctt    420

```

-continued

---

ctgtcacgga aatcaaaatt ggaagtcggg gtttgaatgg aaattttaat ccttcgtact	480
ttcaaaacgc ttttaaaaag cttegaatth ttgatgctag taacaacaac atcgaaggaa	540
atattctca acagtttct acgtctctta ctcaaatgat attgaacaac aataaattga	600
cggaggtct cccacagttt gatcaattgg ggccttgac agtcgtaaac ttgagcaaca	660
acaactctgac cggcaacatg aaccccaact atttcaatgt gatcgtgaat gtggaaacct	720
tcgatgttc ctataacca cttgaaggca ctctcccga ctccattcta aacctggcca	780
agcttcgttt ctgaatttg cagaacaata aatttaatgg taaacttccc gacgatttct	840
ctcggctgaa gaatttgcag actttcaaca ttgagaacga tcagttcacg ggtaattatc	900
catcaggttt acccagtaat agcagggttg gagaaatcg tcttacattt cccccactc	960
cagccccgg caccctgct cccaggactc cttctccttc aggaacatcg aatggatcat	1020
cgtcgcatct cctctaggg gcgatcattg gaatagccgc tgggtgtgct gtgctgcttt	1080
tattactagc actcggcatc tgtttgtgtt gtcgtaagcg gtccaagaaa gcattgggcg	1140
atccagaggc caccgaccgc agccgaagac cgtggttcac acctcccctc tccgcaaaga	1200
gccagagtga tcccagcaag agcatagaca aaacgacgaa acgcaacatc tttggcagca	1260
gtaagagtga gaagaaaagt tcaaagcaca gagtatttga gccagctcct cttgacaaag	1320
gagcagccga cgaaccagtg gtgaaggcgt ctccgcccgt caaggctactg aaggctcctc	1380
cttcatttaa gggatcagc ggcctgggtg ctggacatc gaaagcaaca attggcaagg	1440
tgaacaagag caatattgca gccaccccat tctctgtagc ggatcttcag gcagccaca	1500
acagcttctc ccaggataat ctgattggag aaggagcat gggtcgcgtg tatcgtgccg	1560
agtttccca cggccaggtc ttggccgtga agaagatcga cagcagcgcg tcgatgggtc	1620
agaatgagga tgacttcttg agtgtagtag acagtttggc tcgcctgcag catgctaata	1680
cggctgagct tgtgggttac tgtattgaac atgaccaacg gctgttgggtg tacgagtagc	1740
tgagtcgtgg aaccctgaac gaattgctcc atttctcggg tgaaaacacc aaggccctgt	1800
cctggaatgt ccgcattaag attgctttgg gatccgcgcg tgctctggag tacttgcaacg	1860
aagtctgtgc acctcccgtg gttcaccaca acttcaaate tgccaatatt ctgctagacg	1920
atgagctcaa tctcatggt tcggactgtg gactagctgc ccttgacca tctggttctg	1980
aacgccaggt gtccgacaca atgttgggct ctttcgggta cagtgccctt gagtaccca	2040
tgtctggaac ctataccgtg aagagtgcg tctacagctt cgggtgtgta atgctggagc	2100
tactcactgg gcgcaagtct ttagacagct caagaccacg atccgagcaa tctttggtac	2160
gatgggccc acctcaatg caccgacatc acgcccctgc acgaatgggtg gatccgtcgt	2220
tgaagggcat ctacctgct aaatcactct ctcggtttgc tgatatagtc gccctttgcg	2280
tccagccgga gcccgagttc cgacccccga tgtctgaagt ggtgcaggca cttgtaagge	2340
tgatgcagcg tgcgagctct agcaaacgca gatcggagtc cgtgttggga attgagtcga	2400
acgagccatc tgagacttca ctttgagagt actgaagcgc ccactagcct aatcgtgcat	2460
ctttggccat ctcgtttctg agtgaacac aagctgggta tattctttgg tggtaagca	2520
acattttgtc acaatttgaa cttcagctgg agaaggtct gtagtgttga agaaaacgaa	2580
tgcaaagcgt ttcggcgtgg atgtgctttg agaacttaca aaactcatca agactttgaa	2640
gatctttgta ttgcatcgaa tctttcaat cagtctcggg taggatcagt tctctgtat	2700
cggataccct tttcatccta acatgggacc cttttaatcc agaggatgga gtgcttgaa	2760
tagtgacctt ggtcaggtta acgc	2784

-continued

<210> SEQ ID NO 15  
 <211> LENGTH: 1088  
 <212> TYPE: DNA  
 <213> ORGANISM: *Physcomitrella patens*

<400> SEQUENCE: 15

```

atcccgggag tgggtggtg gactgtaagg agctagcgtt ttagagctac agtgcggttt    60
gctgtgtgag tgagtgagtg agtgagtgcg tgagtgagga tgtctgtttc tggtatggac    120
aactatgaga agctggagaa ggtaggagag gggacttacg gaaaggtgta taaggcccgt    180
gataaacgct ccgggagcgt ggtggcgctc aagaagacta ggttggagat ggaggaagaa    240
ggcgtccctt ccaccgcttt gcgcgaagtt tcggtgctac aaatgctctc ccacagcatg    300
tatatcgtca ggctactttg cgtggagcac gtcgagaaag gcagcaagcc catgctctac    360
ttggtctttg aatatatgga cactgatctt aagaagtata ttgacttgca cggtcgtggt    420
ccgagcggga agcctctgcc tcccaaagtg gtccagagtt tcattgatca attgtgcaca    480
gggcttgccc actgtcatgg ccacggagta atgcacaggg atctgaaacc ccagaatttg    540
ctcgtcgaca agcaaacccg tcgtcttaag attgccgacc ttggtctcgg tcgggcattc    600
acagtgccaa tgaagagtta cacacacgag attgttactc tatggtaacc agctcctgaa    660
gttcttcttg gagcgaccca ctactctcta cctgtggata tctggtctgt tgggtgcac    720
ttcgtgaac tcgtccggaa aatgccgctc ttcactggag actccgaact tcagcagctt    780
cttcacatct tcaggttgct tggcaccocg aatgagacaa tctggcctgg tgttagccag    840
cacctgtgatt ggcacagatt tcctcaatgg agaccacaag atctgtccct tgctgttccc    900
ggactcagcg cggttggctt agacctctc gccaaaatgt tggatttcga gccctcaaag    960
agaatctctg ccaaagccgc cttgagccat acttatttcg ctgatgtga taagacagca   1020
acctaaacac aacagaacaa ttcaagagaa ccaggtaacc tctacctgtc caagacgaag   1080
gttaacgc                                         1088

```

<210> SEQ ID NO 16  
 <211> LENGTH: 1627  
 <212> TYPE: DNA  
 <213> ORGANISM: *Physcomitrella patens*

<400> SEQUENCE: 16

```

atcccgggca acgagaagca ttcgagatgg cagatgcgaa ggaggaactg gcgctgcgca    60
cggaaatgca ctgggctgtg aggagtaacg acgtggggct gttaaggacc attctgaaga    120
aagacaagca gctcgtgaat gctgcggact atgacaagcg cacgcccttg cacatcgccg    180
cgtccctgga ttgtgtccct gttgctaaag tcctgcttgc ggaaggagca gagttgaatg    240
caaaagacag gtgggggaaa tctccgagag gcgagggcga gagtgcagga tacatggaga    300
tggtaaagct gttgaaggat tacggggctg agtcacacgc aggtgccccc agggggccacg    360
ttgagagtct gattcaggtt gcccctccgt tgccttctaa ccgcgactgg gagatcgctc    420
cgtcggagat tgaacttgat accagcagc tcactcggca aggcgccttt ggagagattc    480
ggaaggcgct ttggcgcggc acaccgctc ctgtgaagac aatcagacct tctctgtcca    540
acgacagaat ggtcatcaag gacttccagc acgaggtgca attgctcgta aagggttcggc    600
acccaaacat tgtgcagttc ctcggggctg ttaccctgca aagacctctc atgttagtca    660
ccgagtttct gccagggggc gatttgcatc agttgctgag gagcaacct aatttggtc    720
ctgaccgcat cgtgaagtat gccctcgaca tagctcggg catgtcttac cttcacaatc    780

```



-continued

---

ggagcaagcc catcatccac cgcgatctca aacccccgaaa catcatagtg gacgaagagc	840
atgagctgaa ggtcggcgac ttcggactga gcaagctgat cgacgtaaag cttatgcatg	900
atgtgtacaa gatgacgggg gggactggga gttacagata catggcgect gaggtcttcg	960
aacatcaacc ctacgacaaa tccgtcgacg tgttttcctt tggaatgata ttatatgaga	1020
tgtttgaagg cgtcgcctcg tttgaggaca aggatgcata cgacgctgcc aactagttg	1080
ctagagacga taagcgccca gagatgagag cccaaacgta tccccacaa atgaaggcat	1140
tgatcgagga ttgctggtca ccgtataccc cgaagcgacc accttcgtc gaaatcgta	1200
aaaaactcga ggtaatgat gaggattgct tattgagatt gcccaaagac cgtcgtcatc	1260
tccgacat cttgcatctt cgacgcaatc ctgcagactc gtgattgatc gggccaacct	1320
tcgagctgat caatctaagt agtcaatgcc ttactgtgtc aaattcagcc tccgccgaca	1380
gattggctat ggttcaagtg attggattct ctgcttctcc agagccagaa acgacccccg	1440
tgcaatttct tctccgacga ccacattgag acatgaagca ccagactttg gatgtagaag	1500
gcatggtcta catgctttgc tgtgagcctt gcacgtctcg caggttgatc tctttaacca	1560
gcttctagcc tttcgcaatg gctgcatcac ttaagaaatc accgagtatc gtgatgctcg	1620
ttaacgc	1627

&lt;210&gt; SEQ ID NO 17

&lt;211&gt; LENGTH: 1441

&lt;212&gt; TYPE: DNA

&lt;213&gt; ORGANISM: Physcomitrella patens

&lt;400&gt; SEQUENCE: 17

atcccgggct gtgatgtcgg tgtgggtgctc tgcaagaaat cagatgacgt cataagcatg	60
aaaaggtacc agagacgtaa agttcagaga ctcggctcggg agggccaagt cctattggag	120
agaactcttt tcaagcaact gaggccttcc ccattcgtgc cgcattctctt ggccaccct	180
attgacagtg acaatgtggc actcgttctt aattgtgtgt tagctgggccc tctagaactt	240
ctacttcggt cacctttaga cgagaactca gctcgttttc tggtcgcaa cgtggtatta	300
gccgtcgaac ttctgcacaa ggatggcgtt gtatatcgtg gcattctctc cgatgttctt	360
atgatagatc ggaaaggacg acttcagctg gttgattttc ggtttgcaa gcaaatgtcg	420
gatgagcgca ctttcacagt ctgtggcatg gctgatttct tagcaccoga gatcattcaa	480
ggacaaggtc atggcctggc ttctgactgg tgggcggtag gtgtgtaaat gtacttcatg	540
ttgcaaaactg agcttccatt tggatcatgg cgggacaacg agcttgaaat ttttgtaga	600
atagcccgtc ggcagcttac gtttccttca agtttcagcc ctgaagcggg tgacctcatt	660
gacaagctgc tgggtgtgga cccaaccaag agactgggct gtgacagcca tggatcgctt	720
gccataaggg aacatccttg gttccgaggt ataaactggg acaagcacct cgattgcagt	780
gtggaagttc cttcagagat catgacacgc cttcagttgg ccatagactt tcttcccgtg	840
gatgatagtt atcaagtgtt tgatctccaa cccgatgaag acgatccacc atggcttgat	900
ggctgggtgat agcttgatgg ctcgtagatc ccccttctcc aagcatcaat ggcacagtac	960
cgaatggcta taacagaaga tgcacattaa gtgctccatg aacagatacc gtagcgctta	1020
ggatttttcg gtcgtcacia atgacggctc tcttgtgagg ttcgaatgtt gtgtcacccg	1080
atgatctcta ctggcaciaa cctccaggct gaatcttaag gccagctggt ttaggtgaga	1140
cgtttacctt ggttcgaact cacgctcgtg ttgttaagcg cgagtcgatg atgtatgaaa	1200
tgacggtggt ccttgaaagt cttgaaagc aatcaattcg cttatgtgtg tcccttccat	1260

-continued

---

gtggtcatta ggaagggaa cgcctgcact agtcagtaa cgaacatggc tcaattgta	1320
tagcatagcg gtagaggttt cgtacgaaat gtggttgag tcggtgatta taggcgcatt	1380
tctctgaaca tgcacgagaa tcgtctcct gagtctccat cattcagtgg tgcgagctcg	1440
c	1441

<210> SEQ ID NO 18  
 <211> LENGTH: 1736  
 <212> TYPE: DNA  
 <213> ORGANISM: Physcomitrella patens

<400> SEQUENCE: 18

atcccgggct cacgtagtgc actgaactct gtctgaattt taggggatga gaggtagatt	60
tgaagaatac tgggtctctaa ttttctgta attttccacc cttgaggtag ctcattggatt	120
tgggaggtga tcgcatgaga gctcctcaga ggcagtctcg agaatatcaa tatagatcat	180
tggacgtctt cacagagcag cagcagcagt tgcaaaagca gcagcagcaa gatgagtatc	240
agagaacaga attgaagctc gagacactgc caaaaatggt aagcaatgcg accgtgtcat	300
cttcccctcg aagcagtcgg gatggacgta gactacgtac agtcgcgaat aagtatgctg	360
tggaaggtat ggttgggagt ggcgcattct gcaaggtgta tcagggctcc gatttgacga	420
accacgaggt tgtgggcacg aagctggagg atacgagaac tgagcacgct cagttaatgc	480
acgagtgcgg cttgtacaac atattgctgg gtgggaaggg agtgcccaac atgagatggt	540
tcggaaaaga gcaagactac aatgtgatgg tgctagacct attggggcgg aacctgttgc	600
acctctttaa ggtgtgtggg ctaaggtttt cgttgaagac cgtgattatg ctcggttacc	660
aaatgattga ccgggtggaa tacgtgcatt ctcgagggct cgttcaccgt gacctgaagc	720
cggataactt cctcatgggc tgcggtcggc aaggaaacca agtgttcatt atagattttg	780
gcttggcaaa ggagtacatg gacccggcaa cacgaaggca tatcccttac cgagatagga	840
agagcttcac agggacggca cggtaogcta gtaggaatca gcacagagga atcgagcaca	900
gtagaagaga tgacatagaa tcaacttggt acattcttat gtactttcta agaggcaatt	960
tgccatggca agggaagggc gggcaacgcc tcaactgacca gaagcaacac gagtacatgc	1020
acaacaaaat caagatgaac accactgtgg aggagctttg tgatgggtat cccagtcaat	1080
ttgcccactt tttgcaccac gcgcgaagtc taggtttcta cagcagcct gactactgtt	1140
acctccgaag cttgttcctg gatcttttca ttcagaaaaa attccagctc gaccatgtgt	1200
acgactggac tgtgtatact caactcccc agaatggctc tctgcaatca gtgcgcagcc	1260
agaattccgc tgcctcgtcc catttgcaaa atcgacctc gaatgtatca tattgtccac	1320
ccttgaccaa gtcggagtcc cgtcgtgagg ttgttgccgc gaattagggc ttacgttggg	1380
aggactagtg gttcatcctc tgctctggtc ctaaaatagc acaaggttgc ttactgttcc	1440
cctctctcaa gtcttacatg attgtgaatg ggggtttatg gaggttgagga tgaggcaact	1500
aagcagagtg taggaaaaga gttgtagact ctctagtgtg tagtgtgtaa atcaaggctt	1560
ctagcattgt gtcggtagct tgtatggatc agactagaaa tgactttatc cattacaaga	1620
atctttactc ggaagccca tgacggtgat gatttcaata cgttgcaaaa gcaactttct	1680
tctgtaattg aaatagagga tctggtctga gtatgagaag atgggcatgt taacgc	1736

<210> SEQ ID NO 19  
 <211> LENGTH: 1900  
 <212> TYPE: DNA  
 <213> ORGANISM: Physcomitrella patens

-continued

&lt;400&gt; SEQUENCE: 19

```

atcccgggtt gtcgaggacg gagagagaag agagagagag agagagagag aggtgttgtt    60
taggggagcg atgcccggagc aggatgtgtg ttaagttcgt aaggagaagg gagtacatgc    120
aagtgcgtgc ttgtcggata tcggacagct ggatttghta ataagcggag aggagggtcg    180
gtaatcaggg gcgtacatcg atggagccgc gtgtgggaaa caagtatcgg ctgggacgga    240
aaattgggag cggttccttt ggggagatct atcttgggac caatgttcag accaatgagg    300
aggtcggaat aaagctggaa agcatcaaga cgaagcatcc acaattgctg tacgagtcca    360
agctctaccg gatactacaa ggaggaactg ggattcccaa tatcagatgg ttcgggatag    420
aaggagacta caatgtcttg gttctggatc tgttggggcc aagtctcga gaccttttca    480
acttctgcag ccggaagttc tctttaaaga ctgttctcat gcttgctgac cagctgatca    540
acagagtgga gtatgtgcat gcgaaaagct ttcttcatag agacatcaag cctgataatt    600
ttctaattgg gcttggttagg cgagcaaacc aggtctacat tattgatttt ggtcttgcca    660
agaagtaccg cgacccttc acgcatcagc atattcccta cagggagaac aaaaatctga    720
cagggactgc tcggtatgca agcatcaaca ctcatcttgg tattgagcaa agcagacgag    780
atgatttggg atctcttggg tatgtgctca tgtacttctc gagaggcagt cttccatggc    840
aaggactgaa agcgggaacc aagaagcaga agtacgagaa gatcagtgag aaaaaaatgt    900
ccacgcccat tgaggtcctt tgtaaaaatt atccttcaga attcgctcgc tacttccaact    960
actgccggtc cttgctgttt gatgacaaac ccgactatgc atatttgaag agaactcttc   1020
gtgacctctt tattcgtgag ggttttcaat ttgactacgt tttgactgg acaattctga   1080
agtaccagca gtcacaaatt tccggtggca gttcaactcg actgggtgct tctgcagggc   1140
aaaccagtgg tgcacttggg actgggggta caggaagccg agacctgcag cggcccaccg   1200
aaccaatgga tccttctcgg cgcaggcttc ctggaggagc aaatggctcc ggggtcgcaa   1260
atgctttgga ctcatcctaag cacaaaagtc ctggacttga tgaatctgct aaggattctg   1320
ctcttgctgt tgtgtcagaa ccagagcgca tgcatacatc ttcgtatgca actcgggggg   1380
gttcttctc caggcgagct gtcctatcta gcagcaggcc ctcaggggca tcagcagaag   1440
tcgtagatc ctctcgaaca gggagcagta agcttggctc caccagctta cggctcgtcag   1500
cagggatgca gaggagctct ccagttactt cggacccaaa gcggatatct agccgccatc   1560
cacaaccgcc atctgccaac ttgaggattt acgaagccgc tatcaaggga gttgaatcac   1620
tttctgttga ggtgatcaa agccgttata agtaggccc ggcttgtggt tatatagccg   1680
ggctctgtct tctatcaaac cctcttgta tgtagatgag agttgctcta catttgcaa   1740
cagcctgatt gaggggaaaa cgggtggttc gtcctacaat ggtgctaaga ctacaggtct   1800
ctcactacta ggaatgaatg gatctctatc ttgttaccat caaaccattg tcagtgcctt   1860
gtgtggtagc tctctgcat acgattccta aggttaacgc                               1900

```

&lt;210&gt; SEQ ID NO 20

&lt;211&gt; LENGTH: 1217

&lt;212&gt; TYPE: DNA

&lt;213&gt; ORGANISM: Physcomitrella patens

&lt;400&gt; SEQUENCE: 20

```

gcgttaacgg gaggaaggtc gggggaagag acgcttgagg ctgctgaaag gggattcact    60
cagcgtcccc acccattcgt caatctggcg cagaagatcg gaaaatcggc ccgacggcca    120
gggttatatg ccaagcccg gggttacaca gatgtgaatg tccaacgtcc gaaagattat    180

```

-continued

tgggactacg aggccctcac cgtccaatgg ggggaccaag acgattacga ggtagtgcgt	240
aaggtggggc gagggaaata cagtgaggtt tttgaagggtg tcaacgcogt gaatagtgag	300
cgttgcggtta tgaagattht gaagccagta aagaaaaaaa agatcaaaag agagatcaag	360
attctgcaaa acctttgtgg agggcccaac attgtgaagc ttctggacat tgtccgtgat	420
cagcaatcga agacaccag cctaattttt gagtatgtga acaatactga tttcaaagt	480
ctctacccca ctcttacaga ctttgatcgc cgatactaca ttcattgagct gctcaaggct	540
ttggactatt gccattctca agggattatg cacagggatg tgaagccaca caacgtgatg	600
attgaccatg agcagcggaa gcttaggctt attgactggg gacttgccga attctatcat	660
cctggcaaa agtataatgt gcgtgttgcc tctaggtact tcaagggtcc tgagctgctg	720
gttgatcttc aagattatga ttactctctc gacatgtgga gcttgggggtg catgtttgcc	780
ggcatgatat ttccgaagga gccattcttt tatgggcagc acaattatga tcaactgtg	840
aagattgcta aggtgttggg aactgatgaa ttgaattcct atctaataca ataccgcta	900
gagctggacc cccatttggg agcactggtt ggcaggcata gcaggaaacc ttggtcaaa	960
ttcatcaatg ctgataatca gcgtctggtt gttccagagg ctgtggattt tttggataag	1020
cttctacgct acgatcatca agacaggctg actgcgaagg aagctatggc acatccctat	1080
ttttatcccg tgaaggtgtc ggaggttagc aaccgtcgca gtgcttgata tgaattgata	1140
tatctcatat gggctttctt gtgattacgt cccaccgggc taccagggtt ctcagttgtg	1200
cgaagcgctg agctcgc	1217

&lt;210&gt; SEQ ID NO 21

&lt;211&gt; LENGTH: 1718

&lt;212&gt; TYPE: DNA

&lt;213&gt; ORGANISM: Physcomitrella patens

&lt;400&gt; SEQUENCE: 21

atcccgggag agccatggag ccaactgctt cggcgaatgg gactgtttga cttcttctct	60
tcgccccccg ctgcctcttc accctctctc gttcttctca cagcctctct ctcctctct	120
gtctgtttgc tgggtaagtt ttgggagtga ggaggacgtg gtcattggaag aagagcccc	180
ctcttttcta gtggactgtc ggtaaattgg acctggagcc tgcctgctca tgcctttgc	240
ttagattgtg ggcgggtgct gttgaaattc cttgaaactg ctactggtcg gaaacgctcg	300
aattgcgact ttgattgaa gtctggttgc tgctgcggtc gggatcttac tcagtctctt	360
caataggacc tctgaagcag tatggagact agcagtgga ctcagaatt gaaagttata	420
agtactccga cctacggagg tcattacgtg aaatatgttg tggcgggaac tgatttcgaa	480
gtcaccgcca ggtacaagcc accactctct cggattgggc gcggagctta tggaaatctc	540
tgttcactct ttgataaccg tacgggtgag gaggtggcgg tcaaaaagat tggaaacgcc	600
ttcgacaaca ggatcgatgc gaagcgaaca ctgcgtgaaa taaaactcct cggcatatg	660
gatcatgaaa acgtcgttgc cattacagac atcattctgc cccaactag ggagaatttc	720
aacgacgtgt acattgtata cgagttgatg gatacggacc tacaccagat cattcttca	780
aatcaagctc tcacagaaga ccaactgtcag tattttctgt atcaaatctt gcggggcttg	840
aagtacatcc attcggcga cgtcttgcac cgggacttga agcccaccaa ccttctctgc	900
aatgccaat gcgattttaa aatcgagat tttggcttgg cacgcactct cctgaaacg	960
gatttcatga ctgagatgt tgtaacgagg tggtagag ctccagagct gctcctgaat	1020
tgttcagcat acactgcagc tattgacatt tggctctgtg ggtgcactct catggagt	1080

-continued

---

cttaaccgat	ctgcgttggt	ccctgggaga	gactatgtgc	atcagctccg	cctaattaca	1140
gaactcatcg	gaactcctga	agatagggat	cttgggtttt	tgagaagcga	caatgctagg	1200
cggtatatca	agcacctgcc	tcgacagtcg	cctattccct	taaccagaa	gttcagaggc	1260
attaatcgtt	ctgctcttga	tcttgttgaa	aagatgctgg	tctttgatcc	agcgaaaaga	1320
atcacagtgg	aagctgcctt	ggcgcacct	tatttagctt	cacttcatga	catcaacgat	1380
gagctgcct	cggtatctcc	cttcgagttt	gacttcgagg	agccccctat	ctcggaggag	1440
catatcaagg	atctcatttg	gagggagct	ctggattgca	gcttaggtcc	tgatgatatg	1500
gtgcagtaac	ttcacacttc	atctcaagtt	gtaaggceta	ctctcaattc	tttaggtggc	1560
tacaacgcta	tcccgcggt	gtatggtttt	gcaacttatt	ccccccgtg	tgattacact	1620
attggattat	agaatgacaa	ttcgttagtt	ctttccctg	gcgctatc	ttgtctgca	1680
catttcatcc	agcagacatt	gttgcctggc	gtaaacgc			1718

&lt;210&gt; SEQ ID NO 22

&lt;211&gt; LENGTH: 2177

&lt;212&gt; TYPE: DNA

<213> ORGANISM: *Physcomitrella patens*

&lt;400&gt; SEQUENCE: 22

atcccggtt	tgattggct	cggataattt	atggtgacaa	tgatttggtg	aggcttcgta	60
ttgagtcage	gagcaggtcg	agagttcggc	agcgaagtta	cactcgacct	ggctgaaatt	120
tggaattgaa	gcgctgaa	cttcatctgt	gattttggag	gttgtttgac	tgatgagaag	180
aggctctctga	gctgagaatg	tttgcaattt	aggggcacca	ccggtttggt	ggagtccctt	240
gccacttatt	acaattggtg	gtttacaagc	tcgacgagtt	tcaatcgaac	gtagagtttt	300
agtcgggtcg	aggatctatg	tatccgctca	gcggagaaga	gagcctgatg	tgccgaagc	360
gatcgtgtgg	gatttgacta	gaaagaggtg	gaccgcatca	gaactattta	ttccttgatg	420
gggaaggatc	gaggttccaa	tgggtctcac	tccgttttct	tgtgtcacgg	ttcaaggtta	480
tgtccgggtg	gtctaccctg	acggccacct	cgagaatctg	agcaaatctt	gtagcgtgca	540
cgatcttctt	ctgggtaate	cagactacta	tgtctgctgt	agcaccctt	acacaatcac	600
caatcgtatg	gcagcggaa	aggtgctcga	gtatgggtg	acctactctg	tttgcccaac	660
gccaaatgcc	caacttttct	tagaacgtca	gccgaagta	gtacatcgag	gatccaagat	720
tttgccacga	ttttccaaac	atgggggtcca	tgtgcgggag	ttgcgaagcc	cgacgcatgg	780
gagccaacag	tcacggaaag	tttttgatta	tcattcagta	acgatgcagc	agcttgaatc	840
catacgaaac	gagggcccag	agcctcacct	cgctggagac	cgaccatcga	agcaccttaa	900
gctcgttttc	attcggcatt	gcttgcgagc	acttcgactt	cctagaattt	caatagacct	960
aatggaatcg	ccactcccta	atctttccgg	agaggcetta	tcgccgacgg	caactgccaa	1020
agacgagatt	actcagatga	tactaaaaag	tgccgcaagg	tccgaattag	gaatgtatgt	1080
ttcgaagaga	caggaattct	atcttcgaag	agcgcgtagg	cggcgtaagt	ttgcgtggaa	1140
gccggttttg	cagagcatct	ccgagatgaa	gcctgtcatg	gaattccaca	ctccgatggc	1200
ttaccgggat	agtgggtctc	cgccgaagaa	cgctctacc	ccatccttac	ctggcccga	1260
gaacatttca	cgccacgac	aagtgagtg	cccgaagg	agcagtcctc	cgccgaagaa	1320
cgtctacca	cctccccagc	ccgcatttgt	agcgcggact	gcgtcgaagt	attctgctgc	1380
atctcagcaa	gttcaacgaa	atcgaggcaa	cgcgaaatct	ctttatatgg	cgtagtttgt	1440
gtctcgactg	aactcctatc	tattcccca	tcgagataac	tgcatctggt	ggataaattt	1500

-continued

---

```

ctccaacatt tttgctcttc atcctcaagc agctcctcaa tggccagtaa tatggtacga 1560
cattgtgac aactccaatt acgtagcgtt attctgtaac ccacggtcat cgaggatca 1620
aggaatggcg cagtaagcac tgctactttg tgctttgta tcccgttgtg acgagatgc 1680
atgtgcacc gtgcctatca gtgggatttt cttgagcgca gatcttgctt ccgcagttt 1740
tttcataacg ttttggttcg tagggggcct agacgggtact atcaagcaat gagaagtgtg 1800
ctgggtgga tttgacagca atcttttggg ggattgtctt tcctatgtag aacatagcga 1860
ggacacttgc gctggtggg cacatcccat agaacatagt gcttcacttc tgggttgttc 1920
accactagga tcatatgacc ttctcateta ttttcgggct ttgtttcgag ctcatgtacc 1980
atcgactagc gtcactttga ctgcgggtgat aatcgtttgt caatttagtg gagctttgta 2040
gatgatagat gccatttga cagtagcttg gatgctgttt acaagatagc ggcagctaga 2100
agccttaaac ctttagctac catgtattat ttaaacctat atgaagtga cggctgtgca 2160
gatattgccg ttaacgc 2177

```

&lt;210&gt; SEQ ID NO 23

&lt;211&gt; LENGTH: 1731

&lt;212&gt; TYPE: DNA

&lt;213&gt; ORGANISM: Physcomitrella patens

&lt;400&gt; SEQUENCE: 23

```

atcccggcg gtcgagtcgt attaggtgtt gtttcattgt aagggttcgg aagcacgggg 60
cacggcgtat ataccgttcc ccttgaacgt tgatctcacc tttggaagac ctgaattgag 120
tagcgtgcbg aagctgcacg gatccggaag agacgatgag taggagagtg agaaggggag 180
gtcttcgcbg cgcggtgcbg aagcaagaga ctcccgtcag caaatttttg actgccagtg 240
gaactttcca ggatgatgat atcaagctca accacaccgg gcttcgcbg gtctcttcag 300
aacctaacct tctacgcbg acgcagtcta gctcccaga tgggcaactg tcaatagcag 360
acctggagtt agtgcggttc tgggaaaagg gtgcgggtgg aacctgcbg cttgtccggc 420
acaaatggac caatgtcaat tatgcbgca aggcgataca aatgaatgc aacgaaacag 480
tgaggaagca gattgttcag gagctgaaaa tcaaccaagt gacgcaccag cagtgcctt 540
atatcgtgga atgcttccac tcttctacc acaacggcgt catatccatg atcctagagt 600
acatggacag gggctcgttg tccgacatta ttaagcaaca aaagcagata cctgagccgt 660
atctggcctt cattgctagt caagtctga agggattgga atacctcac caagtcaggc 720
acatcataca tcbgcatata aagccctcca acctcctcat caatcacaag ggtgaggtca 780
aaatctctga ttttggtgcb agtgctgtgt tgggtcattc cttggcccag cgagacacgt 840
tcgttgggac ttgcacatat atgtcgcag aacgccttca gggcgttcg tatgcatacg 900
acagtgacct atggagtta ggattgactc ttttgagtg tgcgttgggt accttcccat 960
acaaaccagc tggaatggaa gagggttggc aaaatttctt catcctcatg gaatgtatag 1020
ttaatcaacc ccccgcbgcb gcatcccctg acaaattctc cccgcaattt tgttctttta 1080
ttgaatcctg catccgaaa tgtcccagtg aacgaccatc aactactgat ttacttaaac 1140
atccgttctt gcaaaagtac aacgaggaag agtaccattt gagcaagatt ttgtaactta 1200
aagttagcct cgcattgcbg gcagagactg tcactaccac aagcctgatc caccactgaa 1260
cttcaagggg ctttaccaaa agcatggtcb aactacctcb ccaatccgcb actttctcaa 1320
tgccttttcc ttatatagtc atatgtgcb aagttgagaa cगतatcaaa tcagattgac 1380
ggaaaaaaca tcttcaacgc cgtttcccaa ccttatagaa agtggagttt tctcaatgag 1440

```

-continued

---

```

ccccatttgt cgctgagaac gtgcagctca tgaacaatc cataagtgtg ttaatcgggg 1500
tcttatatta tcatcaccat gctagctttt tatgttacct gcactttttc tttccttatt 1560
gcacagcadc gaacacttct tcgatacca aaacaatatt tccatcttct tcttctttt 1620
ttcacgtctc tgcgacaagg aatttctca cggagatttt tcaacacttt tctcaaatgt 1680
ttttagagtt tttaaactga caattgaaga ggtcggacct accggactcg c 1731

```

```

<210> SEQ ID NO 24
<211> LENGTH: 1407
<212> TYPE: DNA
<213> ORGANISM: Physcomitrella patens

```

```

<400> SEQUENCE: 24

```

```

atcccgggag aggctgatct gatgtacag tttcgtgtgc agctagtctt tagagattcg 60
ggcaacgcac ttgttgaaga tcggaaactt tcaaatcgg tcgagtcgta ttaggtgttg 120
tttcattgta agggttcggc agcacggggc acggcgtata taccgttccc cttgaacgtt 180
gatctcacct ttggaagacc tgaattgagt agcgtgcgga agctgcatcg atccggaaga 240
gacgatgagt aggagagtga gaaggggagg tcttcgcgtc gccgtgccga agcaagagac 300
tcccgtcagc aaatttttga ctgccagtgg aactttccag gatgatgata tcaagctcaa 360
ccacaccggg cttcgcgtcg tctcttcaga acctaacctt cctacgcaga cgcagtctag 420
ctccccagat gggcaactgt caatagcaga cctggagtta gtgcggttct taggaaaggg 480
tgcgggtgga accgtgcagc ttgtccggca caaatggacc aatgtcaatt atgcactgaa 540
ggcgatacaa atgaatatca acgaaacagt gaggaagcag attgttcagg agctgaaaat 600
caaccaagtg acgcaccagc agtgccctta tatcgtggaa tgcttccact ccttctacca 660
caacggcgtc atatccatga tcctagagta catggacagg ggctcgttgt ccgacattat 720
taagcaacaa aagcagatgc ctgagccgta tctggccgtc attgctagtc aagttctgaa 780
gggattggaa tacctacacc aagtcaggca catcatacat cgtgatataa agccctccaa 840
cctcctcatc aatcacaaag gtgaggtcaa aatatctgat tttggtgtca gtgctgtgtt 900
ggttcattcc ttggcccagc gagacacgtt cgttgggact tgcacatata tctcgcaga 960
acgccttcag gggcgttcgt atgcatacga cagtgaacta tggagttag gattgactct 1020
tttgagtggt gcgttgggta ccttcccata caaacagct ggaatggaag agggttggca 1080
aaatctcttc atcctcatgg aatgtatagt taatcaacc cccgcagccg catccctga 1140
caaatctccc cccgaathtt gttcttttat tgaatcctgc atccggaaat gtcccagtga 1200
acgaccatca actactgatt tacttaacaa tccgttctcg caaagtaca acgaggaaga 1260
gtaccatttg agcaagattt tgtaacctaa agttagcctc gcattggcgtg cagagactgt 1320
cactaccaca agcctgatcc accactgaac ttcaaggagc tttaccaaaa gcattggtcga 1380
actacctcgc caatccgcca gagctca 1407

```

```

<210> SEQ ID NO 25
<211> LENGTH: 2253
<212> TYPE: DNA
<213> ORGANISM: Physcomitrella patens

```

```

<400> SEQUENCE: 25

```

```

atcccgggtg taggcggggc aggttcgatg caatggggca gtgttatgga aagtttgatg 60
atggaggcga agggagggat ttgttgagc ggcagaaagt gcaggtttct aggacccaa 120
agcatggatc gtggagcaat agcaaccgag ggagcttcaa caatggcggg ggggcctcgc 180

```

-continued

---

```

ctatgagagc caagacgtcg ttcgggagca gccatccgtc cccgcggcat cctcagcta 240
gtccgctccc tcaactacag agctcccag cgccttcgac cccgcgacgg aacattttca 300
aaaggccttt tctcctcct tctcccgcga agcacattca gtccagtctc gtgaaacggc 360
atggcgcgaa gccgaaagaa ggagggcgca tccctgaggc tgcgatggt gagaagccct 420
tgataagca tttcggtat cacaagaact tcgctactaa gtatgagctg gggcatgaag 480
tcggtcgcgg gcacttcggt cacacatggt acgcgaaagt acggaaggc gagcataagg 540
gacaagccgt ggcaagtaag ataatctcga aagcgaagat gacgactgct attgcgatcg 600
aggacgtggg acgagaagt aaaatthtga aggctctgac gggacaccag aatttggttc 660
gattctacga tctctcgcag gaccatctaa atgtgtacat tgttatggaa ttatgtgaag 720
gaggtgaatt attggatcga atthtgtctc ggggagggaa gtactcggag gaagacgcca 780
aggttgttgt gcggcagatt ttgagcgttg ttgcgttttg tcacctgcaa ggcgttgttc 840
accgagatct taagcctgag aatthtctgt ttaccacgaa ggatgaatat gctcagctta 900
aggccattga tthtggattg tcagatttca tcaaaccgca tgaagactg aacgatatcg 960
ttggaagcgc atactacggt gcgccggagg tattgcatag gttatattca atggaagctg 1020
acgtatggag cattggagtc atcacgtaca tthtgttatg tggtagtoga cgtthtggg 1080
cgcggaaccga gtcgggcatt tthcgtcgcg tgttgagggc tgacccgagc tthgaagaag 1140
ccccctggcc tccatctct cccgaagcca aggatttcgt gaagcgtctc ctgaataagg 1200
atatgcgaa acgcatgact gctgcacaag cthtaactca tccatggatt cgaagtaaca 1260
acgtgaagat acctctggat atcttagtgt acagacttgt gaggaattat ctctgtgcat 1320
catccatgag aaaggctgct ttgaaggccc tgtcaaagac thtaaccgaa gacgagactt 1380
thtatctacg tactcaattt atgctgctag aaccaagtaa caacggctgt gttacttht 1440
agaatttcag acaggcactg ctgaaaaatt caacagaggc catgaaagag tcacgggttt 1500
ttgaaattct ggaatcgatg gatggtcttc atthcaagaa aatggactth tcagagttct 1560
gtgcagcggc cattagtgtt ctccagttag aagccacaga acgatgggag cagcatgctc 1620
gcgcagctta cgacatattt gagaaagagg gtaaccgagt cattatctct gatgaacttg 1680
cgaagagat gggactagca ccaaatgtac cagcccaagt gthtctagat tggattagac 1740
agtctgatgg tcggctgagt ttcactgggt tcaccaagct gctacatgga atthccagcc 1800
gtgctatcaa aaatctccag cagtgttctt ttgcatcgta cagttcggaa tggagttht 1860
aagctcttht agthtcaact ccgtcttcaa ctgctgcttc gcctcgtctc tgagctgtga 1920
tagcgtatct caagcatatg cacaactcgc atthtctgctg aagtgtttg tcacctca 1980
ttagtcgggc ctctggaact ttcacttatt tggattatth atgtagaagt ccagatcaaa 2040
aagcgaagag gaatggctag atattgtcac aagaagtaac atagtcaaat tcaggagcac 2100
ttaagcacac atgagtgctt thttattgga attcttagat atggaactga tgtthccaag 2160
ggaaggtct atgagcaga gagtggaaat tatagactgg catatggtta agtgatcatt 2220
ggactgccgt tcaactccgg ttgtcgttaa cgc 2253

```

&lt;210&gt; SEQ ID NO 26

&lt;211&gt; LENGTH: 2230

&lt;212&gt; TYPE: DNA

<213> ORGANISM: *Physcomitrella patens*

&lt;400&gt; SEQUENCE: 26

```

atccggggcg aactgcgacg tgagattcca acttggaaagg gcctcgcgta agaccggatc 60

```



-continued

tcgtttctta	cgcttttgcg	cctcgcgata	ttgttacatt	gtttcctctg	gttttattcg	120
attccgccte	tgaaaatgtg	aacgggctgc	aagcttggtt	ttggagcaac	gttggagcat	180
tgaagggttg	cgctcgtccc	tgcccattcc	tcgcttctgc	tctggcctat	gtcatgacga	240
cgtaaggag	aggatttgag	ggtttttaa	gtgatataat	cctccccgag	gagatttctg	300
tgagttgatt	aacttgatc	agcgacatgg	ggaacactag	ttcgagggga	tcgaggaagt	360
ccactcggca	ggtgaatcag	ggagtcgggt	ctcaagacac	ccgagagaag	aatgatagcg	420
tcaatccaaa	gacgagacag	ggtggtagcg	ttggcgcaaa	caactatggc	ggaaagccaa	480
gcagtgggtg	tcaggccgga	gaacgatcca	cctctgcgcc	cgctgctctg	ccgaggccga	540
agccagcacc	gaggtcagta	tccggtgttt	tgggtaagcc	gctgtcagat	attcgtcaat	600
cttaccatcct	gggacgggag	cttgcccgag	ggcagttcgg	agtgacttac	ttgtgtactg	660
acaagatgac	gaatgaggcg	tacgcgtgca	agagcatcgc	caaacggaaa	ctgaccagta	720
aggaggatat	cgaggatgtt	aagcgggagg	ttcagattat	gcatcacctg	tcggggacac	780
ccaatatcgt	ggtgttaaag	gatgtgttcg	aggacaagca	ttccgtgcat	cttgtgatgg	840
agctctgtgc	aggtggcgag	ctcttcgatc	gcatcattgc	caaggggcat	tacagtgagc	900
gcgccgctgc	cgatatgtgc	agagtcacgc	tcaatgtggt	gcacagatgc	cactcattag	960
gggtcttcca	tcgggatctc	aagccagaga	atcttctggt	ggccagcaag	gctgaggatg	1020
cgctctgaa	ggccacagac	ttcggctctg	caactttctt	taagccagga	gatgtgttcc	1080
aggatattgt	tggaaatgcg	tattacgtgg	cccctgaagt	tttgaagaga	agttatggtc	1140
ctgaagctga	tgtttggagt	gcaggcgtga	ttgtgtacat	tctgctgtgt	ggtgtacccc	1200
ccttctgggc	tgaaactgag	cagggtatct	ttgacgctgt	gctcaaaggg	cacatagact	1260
tcgaaaacga	tccatggccg	aaaatctcca	acggggctaa	ggatttgggtg	aggaaaatgc	1320
taaacctcaa	cgtgaagata	cgtctgacgg	cacagcaggt	gttgaacat	ccatggatga	1380
aggaagatgg	tgatgctcca	gacgtgccac	tcgacaatgc	ggtgttgacc	agactgaaaa	1440
atctctcagc	cgccaacaag	atgaaaaagc	tggcgctgaa	ggtgattgca	gagagtctgt	1500
cggaggaaga	gatcgtgggg	ttgagggaga	tgttcaaate	catagataca	gacaacagcg	1560
gcacgggtgac	gttcaggagg	cttaaggaag	ggttgctgaa	gcagggctca	aaacttaatg	1620
aatcggacat	caggaacta	atggaagctg	cagatgtcga	tggaaacggc	aagatcgact	1680
tcaacgagtt	catatcggca	acaatgcaca	tgaacaagac	ggagaaagag	gatcaccttt	1740
gggcagcatt	catgcatttc	gacacggaca	atagcgggta	tatcaccatc	gacgagcttc	1800
aggaagcaat	ggagaagaat	ggaatgggag	atcctgagac	catccaagag	atcatcagcg	1860
aggtggacac	agacaacgac	ggaagaatag	actacgacga	gttcgtagcc	atgatgcgca	1920
agggcaatcc	tggcgtgaa	aacggaggaa	cggtgaacaa	gccagacac	aggtagtage	1980
tcctggttgc	caatttgacg	acgggtttgg	caaggcaaca	gtagtgtttg	ttagctttca	2040
gattcaggtt	cggatttgtt	catgcctccc	tttgtctcga	acaatggact	ctaggccttt	2100
ccaatggaaa	agctattcca	acagggtttg	cataacgtgt	agtagaatga	aagcattgcc	2160
tggggggtgt	acagtgcctg	tgatcttgtg	gagttctcgt	aggatggctt	cggttgatc	2220
tcgttaacgc						2230

&lt;210&gt; SEQ ID NO 27

&lt;211&gt; LENGTH: 749

&lt;212&gt; TYPE: PR1

&lt;213&gt; ORGANISM: Physcomitrella patens

-continued

&lt;400&gt; SEQUENCE: 27

Met Gly Val Asp Met Lys Ala Pro Ala Lys Gln Ser Leu Gly Val Gly  
 1 5 10 15  
 Leu Leu Leu Cys Ser Val Val Ile Leu Ser Val Val Ser Ser Val Tyr  
 20 25 30  
 Gly Gln Val Gln Thr Asp Pro Val Asp Thr Thr Gly Leu Ile Ser Met  
 35 40 45  
 Trp Tyr Asp Leu Lys Gln Ser Gln Ser Leu Thr Gly Trp Thr Gln Asn  
 50 55 60  
 Ala Ser Asn Pro Cys Gly Gln Gln Trp Tyr Gly Val Val Cys Asp Gly  
 65 70 75 80  
 Ser Ser Val Thr Glu Ile Lys Ile Gly Ser Arg Gly Leu Asn Gly Asn  
 85 90 95  
 Phe Asn Pro Ser Tyr Phe Gln Asn Ala Phe Lys Lys Leu Arg Ile Phe  
 100 105 110  
 Asp Ala Ser Asn Asn Asn Ile Glu Gly Asn Ile Pro Gln Gln Phe Pro  
 115 120 125  
 Thr Ser Leu Thr Gln Met Ile Leu Asn Asn Asn Lys Leu Thr Gly Gly  
 130 135 140  
 Leu Pro Gln Phe Asp Gln Leu Gly Ala Leu Thr Val Val Asn Leu Ser  
 145 150 155 160  
 Asn Asn Asn Leu Thr Gly Asn Met Asn Pro Asn Tyr Phe Asn Val Ile  
 165 170 175  
 Val Asn Val Glu Thr Phe Asp Val Ser Tyr Asn Gln Leu Glu Gly Thr  
 180 185 190  
 Leu Pro Asp Ser Ile Leu Asn Leu Ala Lys Leu Arg Phe Leu Asn Leu  
 195 200 205  
 Gln Asn Asn Lys Phe Asn Gly Lys Leu Pro Asp Asp Phe Ser Arg Leu  
 210 215 220  
 Lys Asn Leu Gln Thr Phe Asn Ile Glu Asn Asp Gln Phe Thr Gly Asn  
 225 230 235 240  
 Tyr Pro Ser Gly Leu Pro Ser Asn Ser Arg Val Gly Gly Asn Arg Leu  
 245 250 255  
 Thr Phe Pro Pro Pro Ala Pro Gly Thr Pro Ala Pro Arg Thr Pro  
 260 265 270  
 Ser Pro Ser Gly Thr Ser Asn Gly Ser Ser Ser His Leu Pro Leu Gly  
 275 280 285  
 Ala Ile Ile Gly Ile Ala Ala Gly Gly Ala Val Leu Leu Leu Leu  
 290 295 300  
 Ala Leu Gly Ile Cys Leu Cys Cys Arg Lys Arg Ser Lys Lys Ala Leu  
 305 310 315 320  
 Gly Asp Pro Glu Ala Thr Thr Ser Ser Arg Arg Pro Trp Phe Thr Pro  
 325 330 335  
 Pro Leu Ser Ala Lys Ser Gln Ser Asp Pro Ser Lys Ser Ile Asp Lys  
 340 345 350  
 Thr Thr Lys Arg Asn Ile Phe Gly Ser Ser Lys Ser Glu Lys Lys Ser  
 355 360 365  
 Ser Lys His Arg Val Phe Glu Pro Ala Pro Leu Asp Lys Gly Ala Ala  
 370 375 380  
 Asp Glu Pro Val Val Lys Ala Ser Pro Pro Val Lys Val Leu Lys Ala  
 385 390 395 400  
 Pro Pro Ser Phe Lys Gly Ile Ser Gly Leu Gly Ala Gly His Ser Lys  
 405 410 415

-continued

---

Ala Thr Ile Gly Lys Val Asn Lys Ser Asn Ile Ala Ala Thr Pro Phe  
420 425 430

Ser Val Ala Asp Leu Gln Ala Ala Thr Asn Ser Phe Ser Gln Asp Asn  
435 440 445

Leu Ile Gly Glu Gly Ser Met Gly Arg Val Tyr Arg Ala Glu Phe Pro  
450 455 460

Asn Gly Gln Val Leu Ala Val Lys Lys Ile Asp Ser Ser Ala Ser Met  
465 470 475 480

Val Gln Asn Glu Asp Asp Phe Leu Ser Val Val Asp Ser Leu Ala Arg  
485 490 495

Leu Gln His Ala Asn Thr Ala Glu Leu Val Gly Tyr Cys Ile Glu His  
500 505 510

Asp Gln Arg Leu Leu Val Tyr Glu Tyr Val Ser Arg Gly Thr Leu Asn  
515 520 525

Glu Leu Leu His Phe Ser Gly Glu Asn Thr Lys Ala Leu Ser Trp Asn  
530 535 540

Val Arg Ile Lys Ile Ala Leu Gly Ser Ala Arg Ala Leu Glu Tyr Leu  
545 550 555 560

His Glu Val Cys Ala Pro Pro Val Val His His Asn Phe Lys Ser Ala  
565 570 575

Asn Ile Leu Leu Asp Asp Glu Leu Asn Pro His Val Ser Asp Cys Gly  
580 585 590

Leu Ala Ala Leu Ala Pro Ser Gly Ser Glu Arg Gln Val Ser Ala Gln  
595 600 605

Met Leu Gly Ser Phe Gly Tyr Ser Ala Pro Glu Tyr Ala Met Ser Gly  
610 615 620

Thr Tyr Thr Val Lys Ser Asp Val Tyr Ser Phe Gly Val Val Met Leu  
625 630 635 640

Glu Leu Leu Thr Gly Arg Lys Ser Leu Asp Ser Ser Arg Pro Arg Ser  
645 650 655

Glu Gln Ser Leu Val Arg Trp Ala Thr Pro Gln Leu His Asp Ile Asp  
660 665 670

Ala Leu Ala Arg Met Val Asp Pro Ser Leu Lys Gly Ile Tyr Pro Ala  
675 680 685

Lys Ser Leu Ser Arg Phe Ala Asp Ile Val Ala Leu Cys Val Gln Pro  
690 695 700

Glu Pro Glu Phe Arg Pro Pro Met Ser Glu Val Val Gln Ala Leu Val  
705 710 715 720

Arg Leu Met Gln Arg Ala Ser Leu Ser Lys Arg Arg Ser Glu Ser Ala  
725 730 735

Val Gly Ile Glu Ser Asn Glu Pro Ser Glu Thr Ser Leu  
740 745

&lt;210&gt; SEQ ID NO 28

&lt;211&gt; LENGTH: 308

&lt;212&gt; TYPE: PRT

&lt;213&gt; ORGANISM: Physcomitrella patens

&lt;400&gt; SEQUENCE: 28

Met Ser Val Ser Gly Met Asp Asn Tyr Glu Lys Leu Glu Lys Val Gly  
1 5 10 15

Glu Gly Thr Tyr Gly Lys Val Tyr Lys Ala Arg Asp Lys Arg Ser Gly  
20 25 30

Gln Leu Val Ala Leu Lys Lys Thr Arg Leu Glu Met Glu Glu Glu Gly  
35 40 45

-continued

---

Val Pro Ser Thr Ala Leu Arg Glu Val Ser Leu Leu Gln Met Leu Ser  
 50 55 60  
 His Ser Met Tyr Ile Val Arg Leu Leu Cys Val Glu His Val Glu Lys  
 65 70 75 80  
 Gly Ser Lys Pro Met Leu Tyr Leu Val Phe Glu Tyr Met Asp Thr Asp  
 85 90 95  
 Leu Lys Lys Tyr Ile Asp Leu His Gly Arg Gly Pro Ser Gly Lys Pro  
 100 105 110  
 Leu Pro Pro Lys Val Val Gln Ser Phe Met Tyr Gln Leu Cys Thr Gly  
 115 120 125  
 Leu Ala His Cys His Gly His Gly Val Met His Arg Asp Leu Lys Pro  
 130 135 140  
 Gln Asn Leu Leu Val Asp Lys Gln Thr Arg Arg Leu Lys Ile Ala Asp  
 145 150 155 160  
 Leu Gly Leu Gly Arg Ala Phe Thr Val Pro Met Lys Ser Tyr Thr His  
 165 170 175  
 Glu Ile Val Thr Leu Trp Tyr Arg Ala Pro Glu Val Leu Leu Gly Ala  
 180 185 190  
 Thr His Tyr Ser Leu Pro Val Asp Ile Trp Ser Val Gly Cys Ile Phe  
 195 200 205  
 Ala Glu Leu Val Arg Lys Met Pro Leu Phe Thr Gly Asp Ser Glu Leu  
 210 215 220  
 Gln Gln Leu Leu His Ile Phe Arg Leu Leu Gly Thr Pro Asn Glu Thr  
 225 230 235 240  
 Ile Trp Pro Gly Val Ser Gln His Arg Asp Trp His Glu Phe Pro Gln  
 245 250 255  
 Trp Arg Pro Gln Asp Leu Ser Leu Ala Val Pro Gly Leu Ser Ala Val  
 260 265 270  
 Gly Leu Asp Leu Leu Ala Lys Met Leu Val Phe Glu Pro Ser Lys Arg  
 275 280 285  
 Ile Ser Ala Lys Ala Ala Leu Ser His Thr Tyr Phe Ala Asp Val Asp  
 290 295 300  
 Lys Thr Ala Thr  
 305

<210> SEQ ID NO 29  
 <211> LENGTH: 425  
 <212> TYPE: PRT  
 <213> ORGANISM: Physcomitrella patens

<400> SEQUENCE: 29

Met Ala Asp Ala Lys Glu Glu Leu Ala Leu Arg Thr Glu Met His Trp  
 1 5 10 15  
 Ala Val Arg Ser Asn Asp Val Gly Leu Leu Arg Thr Ile Leu Lys Lys  
 20 25 30  
 Asp Lys Gln Leu Val Asn Ala Ala Asp Tyr Asp Lys Arg Thr Pro Leu  
 35 40 45  
 His Ile Ala Ala Ser Leu Asp Cys Val Pro Val Ala Lys Val Leu Leu  
 50 55 60  
 Ala Glu Gly Ala Glu Leu Asn Ala Lys Asp Arg Trp Gly Lys Ser Pro  
 65 70 75 80  
 Arg Gly Glu Ala Glu Ser Ala Gly Tyr Met Glu Met Val Lys Leu Leu  
 85 90 95  
 Lys Asp Tyr Gly Ala Glu Ser His Ala Gly Ala Pro Arg Gly His Val  
 100 105 110

-continued

Glu Ser Leu Ile Gln Val Ala Pro Pro Leu Pro Ser Asn Arg Asp Trp  
 115 120 125  
 Glu Ile Ala Pro Ser Glu Ile Glu Leu Asp Thr Ser Glu Leu Ile Gly  
 130 135 140  
 Lys Gly Ala Phe Gly Glu Ile Arg Lys Ala Leu Trp Arg Gly Thr Pro  
 145 150 155 160  
 Val Ala Val Lys Thr Ile Arg Pro Ser Leu Ser Asn Asp Arg Met Val  
 165 170 175  
 Ile Lys Asp Phe Gln His Glu Val Gln Leu Leu Val Lys Val Arg His  
 180 185 190  
 Pro Asn Ile Val Gln Phe Leu Gly Ala Val Thr Arg Gln Arg Pro Leu  
 195 200 205  
 Met Leu Val Thr Glu Phe Leu Ala Gly Gly Asp Leu His Gln Leu Leu  
 210 215 220  
 Arg Ser Asn Pro Asn Leu Ala Pro Asp Arg Ile Val Lys Tyr Ala Leu  
 225 230 235 240  
 Asp Ile Ala Arg Gly Met Ser Tyr Leu His Asn Arg Ser Lys Pro Ile  
 245 250 255  
 Ile His Arg Asp Leu Lys Pro Arg Asn Ile Ile Val Asp Glu Glu His  
 260 265 270  
 Glu Leu Lys Val Gly Asp Phe Gly Leu Ser Lys Leu Ile Asp Val Lys  
 275 280 285  
 Leu Met His Asp Val Tyr Lys Met Thr Gly Gly Thr Gly Ser Tyr Arg  
 290 295 300  
 Tyr Met Ala Pro Glu Val Phe Glu His Gln Pro Tyr Asp Lys Ser Val  
 305 310 315 320  
 Asp Val Phe Ser Phe Gly Met Ile Leu Tyr Glu Met Phe Glu Gly Val  
 325 330 335  
 Ala Pro Phe Glu Asp Lys Asp Ala Tyr Asp Ala Ala Thr Leu Val Ala  
 340 345 350  
 Arg Asp Asp Lys Arg Pro Glu Met Arg Ala Gln Thr Tyr Pro Pro Gln  
 355 360 365  
 Met Lys Ala Leu Ile Glu Asp Cys Trp Ser Pro Tyr Thr Pro Lys Arg  
 370 375 380  
 Pro Pro Phe Val Glu Ile Val Lys Lys Leu Glu Val Met Tyr Glu Asp  
 385 390 395 400  
 Cys Leu Leu Arg Leu Pro Lys Asp Arg Arg His Leu Arg Asp Ile Leu  
 405 410 415  
 His Leu Arg Arg Asn Pro Ala Asp Ser  
 420 425

<210> SEQ ID NO 30  
 <211> LENGTH: 283  
 <212> TYPE: PRT  
 <213> ORGANISM: Physcomitrella patens

<400> SEQUENCE: 30

Met Lys Arg Tyr Gln Arg Arg Lys Val Gln Arg Leu Gly Arg Glu Gly  
 1 5 10 15  
 Gln Val Leu Leu Glu Arg Thr Leu Phe Lys Gln Leu Arg Pro Ser Pro  
 20 25 30  
 Phe Val Pro His Leu Leu Ala Thr Pro Ile Asp Ser Asp Asn Val Ala  
 35 40 45  
 Leu Val Leu Asn Cys Val Leu Ala Gly Pro Leu Glu Leu Leu Leu Arg  
 50 55 60

-continued

---

Ser Pro Leu Asp Glu Asn Ser Ala Arg Phe Leu Val Ala Asn Val Val  
65 70 75 80

Leu Ala Val Glu Leu Leu His Lys Asp Gly Val Val Tyr Arg Gly Ile  
85 90 95

Ser Pro Asp Val Leu Met Ile Asp Arg Lys Gly Arg Leu Gln Leu Val  
100 105 110

Asp Phe Arg Phe Ala Lys Gln Met Ser Asp Glu Arg Thr Phe Thr Val  
115 120 125

Cys Gly Met Ala Asp Phe Leu Ala Pro Glu Ile Ile Gln Gly Gln Gly  
130 135 140

His Gly Leu Ala Ser Asp Trp Trp Ala Val Gly Val Leu Met Tyr Phe  
145 150 155 160

Met Leu Gln Thr Glu Leu Pro Phe Gly Ser Trp Arg Asp Asn Glu Leu  
165 170 175

Glu Ile Phe Gly Arg Ile Ala Arg Arg Gln Leu Thr Phe Pro Ser Ser  
180 185 190

Phe Ser Pro Glu Ala Val Asp Leu Ile Asp Lys Leu Leu Val Val Asp  
195 200 205

Pro Thr Lys Arg Leu Gly Cys Asp Ser His Gly Ser Leu Ala Ile Arg  
210 215 220

Glu His Pro Trp Phe Arg Gly Ile Asn Trp Asp Lys His Leu Asp Cys  
225 230 235 240

Ser Val Glu Val Pro Ser Glu Ile Met Thr Arg Leu Gln Leu Ala Ile  
245 250 255

Asp Phe Leu Pro Val Asp Asp Ser Tyr Gln Val Phe Asp Leu Gln Pro  
260 265 270

Asp Glu Asp Asp Pro Pro Trp Leu Asp Gly Trp  
275 280

<210> SEQ ID NO 31  
 <211> LENGTH: 417  
 <212> TYPE: PRT  
 <213> ORGANISM: Physcomitrella patens

<400> SEQUENCE: 31

Met Asp Leu Gly Gly Asp Arg Met Arg Ala Pro Gln Arg Gln Ser Arg  
1 5 10 15

Glu Tyr Gln Tyr Arg Ser Leu Asp Val Phe Thr Glu Gln His Glu Gln  
20 25 30

Leu Gln Lys Gln Gln Gln Gln Asp Glu Tyr Gln Arg Thr Glu Leu Lys  
35 40 45

Leu Glu Thr Leu Pro Lys Met Leu Ser Asn Ala Thr Val Ser Ser Ser  
50 55 60

Pro Arg Ser Ser Pro Asp Gly Arg Arg Leu Arg Thr Val Ala Asn Lys  
65 70 75 80

Tyr Ala Val Glu Gly Met Val Gly Ser Gly Ala Phe Cys Lys Val Tyr  
85 90 95

Gln Gly Ser Asp Leu Thr Asn His Glu Val Val Gly Ile Lys Leu Glu  
100 105 110

Asp Thr Arg Thr Glu His Ala Gln Leu Met His Glu Ser Arg Leu Tyr  
115 120 125

Asn Ile Leu Arg Gly Gly Lys Gly Val Pro Asn Met Arg Trp Phe Gly  
130 135 140

Lys Glu Gln Asp Tyr Asn Val Met Val Leu Asp Leu Leu Gly Pro Asn  
145 150 155 160

-continued

Leu Leu His Leu Phe Lys Val Cys Gly Leu Arg Phe Ser Leu Lys Thr  
 165 170 175  
 Val Ile Met Leu Gly Tyr Gln Met Ile Asp Arg Val Glu Tyr Val His  
 180 185 190  
 Ser Arg Gly Leu Val His Arg Asp Leu Lys Pro Asp Asn Phe Leu Met  
 195 200 205  
 Gly Cys Gly Arg Gln Gly Asn Gln Val Phe Ile Ile Asp Phe Gly Leu  
 210 215 220  
 Ala Lys Glu Tyr Met Asp Pro Ala Thr Arg Arg His Ile Pro Tyr Arg  
 225 230 235 240  
 Asp Arg Lys Ser Phe Thr Gly Thr Ala Arg Tyr Ala Ser Arg Asn Gln  
 245 250 255  
 His Arg Gly Ile Glu His Ser Arg Arg Asp Asp Ile Glu Ser Leu Gly  
 260 265 270  
 Tyr Ile Leu Met Tyr Phe Leu Arg Gly Asn Leu Pro Trp Gln Gly Lys  
 275 280 285  
 Gly Gly Gln Arg Leu Thr Asp Gln Lys Gln His Glu Tyr Met His Asn  
 290 295 300  
 Lys Ile Lys Met Asn Thr Thr Val Glu Glu Leu Cys Asp Gly Tyr Pro  
 305 310 315 320  
 Ser Gln Phe Ala Asp Phe Leu His His Ala Arg Ser Leu Gly Phe Tyr  
 325 330 335  
 Glu Gln Pro Asp Tyr Cys Tyr Leu Arg Ser Leu Phe Arg Asp Leu Phe  
 340 345 350  
 Ile Gln Lys Lys Phe Gln Leu Asp His Val Tyr Asp Trp Thr Val Tyr  
 355 360 365  
 Thr Gln Leu Pro Gln Asn Gly Ser Leu Gln Ser Val Arg Ser Gln Asn  
 370 375 380  
 Ser Ala Ala Ser Ser His Leu Gln Asn Arg Pro Ser Asn Val Ser Tyr  
 385 390 395 400  
 Cys Pro Pro Leu Thr Lys Ser Glu Phe Arg Arg Glu Val Val Ala Ala  
 405 410 415

Asn

<210> SEQ ID NO 32  
 <211> LENGTH: 484  
 <212> TYPE: PRT  
 <213> ORGANISM: Physcomitrella patens

&lt;400&gt; SEQUENCE: 32

Met Glu Pro Arg Val Gly Asn Lys Tyr Arg Leu Gly Arg Lys Ile Gly  
 1 5 10 15  
 Ser Gly Ser Phe Gly Glu Ile Tyr Leu Gly Thr Asn Val Gln Thr Asn  
 20 25 30  
 Glu Glu Val Gly Ile Lys Leu Glu Ser Ile Lys Thr Lys His Pro Gln  
 35 40 45  
 Leu Leu Tyr Glu Ser Lys Leu Tyr Arg Ile Leu Gln Gly Gly Thr Gly  
 50 55 60  
 Ile Pro Asn Ile Arg Trp Phe Gly Ile Glu Gly Asp Tyr Asn Val Leu  
 65 70 75 80  
 Val Leu Asp Leu Leu Gly Pro Ser Leu Glu Asp Leu Phe Asn Phe Cys  
 85 90 95  
 Ser Arg Lys Phe Ser Leu Lys Thr Val Leu Met Leu Ala Asp Gln Leu  
 100 105 110  
 Ile Asn Arg Val Glu Tyr Val His Ala Lys Ser Phe Leu His Arg Asp

-continued

115	120	125
Ile Lys Pro Asp Asn Phe Leu Met Gly Leu Gly Arg Arg Ala Asn Gln 130 135 140		
Val Tyr Ile Ile Asp Phe Gly Leu Ala Lys Lys Tyr Arg Asp Pro Ser 145 150 155		
Thr His Gln His Ile Pro Tyr Arg Glu Asn Lys Asn Leu Thr Gly Thr 165 170 175		
Ala Arg Tyr Ala Ser Ile Asn Thr His Leu Gly Ile Glu Gln Ser Arg 180 185 190		
Arg Asp Asp Leu Glu Ser Leu Gly Tyr Val Leu Met Tyr Phe Leu Arg 195 200 205		
Gly Ser Leu Pro Trp Gln Gly Leu Lys Ala Gly Thr Lys Lys Gln Lys 210 215 220		
Tyr Glu Lys Ile Ser Glu Lys Lys Met Ser Thr Pro Ile Glu Val Leu 225 230 235		
Cys Lys Asn Tyr Pro Ser Glu Phe Ala Ser Tyr Phe His Tyr Cys Arg 245 250 255		
Ser Leu Arg Phe Asp Asp Lys Pro Asp Tyr Ala Tyr Leu Lys Arg Ile 260 265 270		
Phe Arg Asp Leu Phe Ile Arg Glu Gly Phe Gln Phe Asp Tyr Val Phe 275 280 285		
Asp Trp Thr Ile Leu Lys Tyr Gln Gln Ser Gln Ile Ser Gly Gly Ser 290 295 300		
Ser Thr Arg Leu Gly Ala Ser Ala Gly Gln Thr Ser Gly Ala Leu Gly 305 310 315		
Thr Gly Ala Thr Gly Ser Arg Asp Leu Gln Arg Pro Thr Glu Pro Met 325 330 335		
Asp Pro Ser Arg Arg Arg Leu Pro Gly Gly Ala Asn Gly Ser Gly Val 340 345 350		
Ala Asn Ala Leu Asp Ser Ser Lys His Lys Ser Pro Gly Leu Asp Glu 355 360 365		
Ser Ala Lys Asp Ser Ala Leu Ala Val Val Ser Glu Pro Glu Arg Met 370 375 380		
His Thr Ser Ser Tyr Ala Thr Arg Gly Gly Ser Ser Ser Arg Arg Ala 385 390 395 400		
Val Leu Ser Ser Ser Arg Pro Ser Gly Ala Ser Ala Glu Val Val Asp 405 410 415		
Ser Ser Arg Thr Gly Ser Ser Lys Leu Gly Pro Thr Ser Leu Arg Ser 420 425 430		
Ser Ala Gly Met Gln Arg Ser Ser Pro Val Thr Ser Asp Pro Lys Arg 435 440 445		
Ile Ser Ser Arg His Pro Gln Pro Pro Ser Ala Asn Leu Arg Ile Tyr 450 455 460		
Glu Ala Ala Ile Lys Gly Val Glu Ser Leu Ser Val Glu Val Asp Gln 465 470 475 480		
Ser Arg Tyr Lys		

&lt;210&gt; SEQ ID NO 33

&lt;211&gt; LENGTH: 333

&lt;212&gt; TYPE: PRT

&lt;213&gt; ORGANISM: Physcomitrella patens

&lt;400&gt; SEQUENCE: 33

Met Ser Lys Ala Arg Val Tyr Thr Asp Val Asn Val Gln Arg Pro Lys 1 5 10 15
--



-continued

Asp Tyr Trp Asp Tyr Glu Ala Leu Thr Val Gln Trp Gly Asp Gln Asp  
                   20                                  25                                  30  
 Asp Tyr Glu Val Val Arg Lys Val Gly Arg Gly Lys Tyr Ser Glu Val  
                   35                                  40                                  45  
 Phe Glu Gly Val Asn Ala Val Asn Ser Glu Arg Cys Val Met Lys Ile  
                   50                                  55                                  60  
 Leu Lys Pro Val Lys Lys Lys Lys Ile Lys Arg Glu Ile Lys Ile Leu  
                   65                                  70                                  75                                  80  
 Gln Asn Leu Cys Gly Gly Pro Asn Ile Val Lys Leu Leu Asp Ile Val  
                                   85                                  90                                  95  
 Arg Asp Gln Gln Ser Lys Thr Pro Ser Leu Ile Phe Glu Tyr Val Asn  
                                   100                                  105                                  110  
 Asn Thr Asp Phe Lys Val Leu Tyr Pro Thr Leu Thr Asp Phe Asp Ile  
                                   115                                  120                                  125  
 Arg Tyr Tyr Ile His Glu Leu Leu Lys Ala Leu Asp Tyr Cys His Ser  
                                   130                                  135                                  140  
 Gln Gly Ile Met His Arg Asp Val Lys Pro His Asn Val Met Ile Asp  
                                   145                                  150                                  155                                  160  
 His Glu Gln Arg Lys Leu Arg Leu Ile Asp Trp Gly Leu Ala Glu Phe  
                                   165                                  170                                  175  
 Tyr His Pro Gly Lys Glu Tyr Asn Val Arg Val Ala Ser Arg Tyr Phe  
                                   180                                  185                                  190  
 Lys Gly Pro Glu Leu Leu Val Asp Leu Gln Asp Tyr Asp Tyr Ser Leu  
                                   195                                  200                                  205  
 Asp Met Trp Ser Leu Gly Cys Met Phe Ala Gly Met Ile Phe Arg Lys  
                                   210                                  215                                  220  
 Glu Pro Phe Phe Tyr Gly His Asp Asn Tyr Asp Gln Leu Val Lys Ile  
                                   225                                  230                                  235                                  240  
 Ala Lys Val Leu Gly Thr Asp Glu Leu Asn Ser Tyr Leu Asn Lys Tyr  
                                   245                                  250                                  255  
 Arg Leu Glu Leu Asp Pro His Leu Glu Ala Leu Val Gly Arg His Ser  
                                   260                                  265                                  270  
 Arg Lys Pro Trp Ser Lys Phe Ile Asn Ala Asp Asn Gln Arg Leu Val  
                                   275                                  280                                  285  
 Val Pro Glu Ala Val Asp Phe Leu Asp Lys Leu Leu Arg Tyr Asp His  
                                   290                                  295                                  300  
 Gln Asp Arg Leu Thr Ala Lys Glu Ala Met Ala His Pro Tyr Phe Tyr  
                                   305                                  310                                  315                                  320  
 Pro Val Lys Val Ser Glu Val Ser Asn Arg Arg Ser Ala  
                                   325                                  330

&lt;210&gt; SEQ ID NO 34

&lt;211&gt; LENGTH: 375

&lt;212&gt; TYPE: PRT

&lt;213&gt; ORGANISM: Physcomitrella patens

&lt;400&gt; SEQUENCE: 34

Met Glu Thr Ser Ser Gly Thr Pro Glu Leu Lys Val Ile Ser Thr Pro  
   1                  5                                  10                                  15  
 Thr Tyr Gly Gly His Tyr Val Lys Tyr Val Val Ala Gly Thr Asp Phe  
                   20                                  25                                  30  
 Glu Val Thr Ala Arg Tyr Lys Pro Pro Leu Arg Pro Ile Gly Arg Gly  
                   35                                  40                                  45  
 Ala Tyr Gly Ile Val Cys Ser Leu Phe Asp Thr Val Thr Gly Glu Glu  
                   50                                  55                                  60

-continued

Val Ala Val Lys Lys Ile Gly Asn Ala Phe Asp Asn Arg Ile Asp Ala  
 65 70 75 80  
 Lys Arg Thr Leu Arg Glu Ile Lys Leu Leu Arg His Met Asp His Glu  
 85 90 95  
 Asn Val Val Ala Ile Thr Asp Ile Ile Arg Pro Pro Thr Arg Glu Asn  
 100 105 110  
 Phe Asn Asp Val Tyr Ile Val Tyr Glu Leu Met Asp Thr Asp Leu His  
 115 120 125  
 Gln Ile Ile Arg Ser Asn Gln Ala Leu Thr Glu Asp His Cys Gln Tyr  
 130 135 140  
 Phe Leu Tyr Gln Ile Leu Arg Gly Leu Lys Tyr Ile His Ser Ala Asn  
 145 150 155 160  
 Val Leu His Arg Asp Leu Lys Pro Thr Asn Leu Leu Val Asn Ala Asn  
 165 170 175  
 Cys Asp Leu Lys Ile Ala Asp Phe Gly Leu Ala Arg Thr Leu Ser Glu  
 180 185 190  
 Thr Asp Phe Met Thr Glu Tyr Val Val Thr Arg Trp Tyr Arg Ala Pro  
 195 200 205  
 Glu Leu Leu Leu Asn Cys Ser Ala Tyr Thr Ala Ala Ile Asp Ile Trp  
 210 215 220  
 Ser Val Gly Cys Ile Phe Met Glu Leu Leu Asn Arg Ser Ala Leu Phe  
 225 230 235 240  
 Pro Gly Arg Asp Tyr Val His Gln Leu Arg Leu Ile Thr Glu Leu Ile  
 245 250 255  
 Gly Thr Pro Glu Asp Arg Asp Leu Gly Phe Leu Arg Ser Asp Asn Ala  
 260 265 270  
 Arg Arg Tyr Ile Lys His Leu Pro Arg Gln Ser Pro Ile Pro Leu Thr  
 275 280 285  
 Gln Lys Phe Arg Gly Ile Asn Arg Ser Ala Leu Asp Leu Val Glu Lys  
 290 295 300  
 Met Leu Val Phe Asp Pro Ala Lys Arg Ile Thr Val Glu Ala Ala Leu  
 305 310 315 320  
 Ala His Pro Tyr Leu Ala Ser Leu His Asp Ile Asn Asp Glu Pro Ala  
 325 330 335  
 Ser Val Ser Pro Phe Glu Phe Asp Phe Glu Glu Pro Pro Ile Ser Glu  
 340 345 350  
 Glu His Ile Lys Asp Leu Ile Trp Arg Glu Ala Leu Asp Cys Ser Leu  
 355 360 365  
 Gly Pro Asp Asp Met Val Gln  
 370 375

&lt;210&gt; SEQ ID NO 35

&lt;211&gt; LENGTH: 331

&lt;212&gt; TYPE: PRT

&lt;213&gt; ORGANISM: Physcomitrella patens

&lt;400&gt; SEQUENCE: 35

Met Gly Leu Thr Pro Phe Ser Cys Val Thr Val Gln Gly Tyr Val Arg  
 1 5 10 15  
 Val Val Tyr Pro Asp Gly His Val Glu Asn Leu Ser Lys Ser Cys Ser  
 20 25 30  
 Val His Asp Leu Leu Leu Gly Asn Pro Asp Tyr Tyr Val Cys Gly Ser  
 35 40 45  
 Thr Pro Tyr Thr Ile Thr Asn Arg Met Ala Ala Glu Glu Val Leu Glu  
 50 55 60

-continued

---

Tyr Gly Val Thr Tyr Phe Val Cys Ala Thr Pro Asn Ala Gln Pro Phe  
 65 70 75 80  
 Leu Glu Arg Gln Pro Lys Val Val His Arg Gly Ser Lys Ile Leu Pro  
 85 90 95  
 Arg Phe Ser Lys His Gly Val His Val Arg Glu Leu Arg Ser Pro Thr  
 100 105 110  
 His Gly Ser Gln Gln Ser Arg Lys Val Phe Asp Tyr His Ser Val Thr  
 115 120 125  
 Met Gln Gln Leu Glu Ser Ile Arg Asn Glu Gly Pro Glu Pro His Leu  
 130 135 140  
 Ala Gly Asp Arg Pro Ser Lys His Leu Lys Leu Val Phe Ile Arg His  
 145 150 155 160  
 Cys Leu Arg Ala Leu Arg Leu Pro Arg Ile Ser Ile Asp Leu Met Glu  
 165 170 175  
 Ser Pro Leu Pro Asn Leu Ser Gly Glu Ala Leu Ser Pro Thr Ala Thr  
 180 185 190  
 Ala Lys Asp Glu Ile Thr Gln Met Ile Leu Lys Ser Ala Ala Arg Ser  
 195 200 205  
 Glu Leu Gly Met Tyr Val Ser Lys Arg Gln Glu Phe Tyr Leu Arg Arg  
 210 215 220  
 Ala Arg Arg Arg Arg Lys Phe Ala Trp Lys Pro Val Leu Gln Ser Ile  
 225 230 235 240  
 Ser Glu Met Lys Pro Val Met Glu Phe His Thr Pro Met Ala Tyr Arg  
 245 250 255  
 Asp Ser Gly Ser Pro Pro Lys Asn Ala Ser Thr Pro Ser Leu Pro Gly  
 260 265 270  
 Pro Lys Asn Ile Ser Pro Pro Arg Gln Val Ser Val Pro Gln Arg Ser  
 275 280 285  
 Ser Pro Pro Pro Lys Asn Val Ser Pro Pro Pro Gln Pro Ala Phe Val  
 290 295 300  
 Ala Arg Thr Ala Ser Lys Tyr Ser Ala Ala Ser Gln Gln Val Gln Arg  
 305 310 315 320  
 Asn Arg Gly Asn Ala Lys Ser Leu Tyr Met Ala  
 325 330

&lt;210&gt; SEQ ID NO 36

&lt;211&gt; LENGTH: 346

&lt;212&gt; TYPE: PRT

&lt;213&gt; ORGANISM: Physcomitrella patens

&lt;400&gt; SEQUENCE: 36

Met Ser Arg Arg Val Arg Arg Gly Gly Leu Arg Val Ala Val Pro Lys  
 1 5 10 15  
 Gln Glu Thr Pro Val Ser Lys Phe Leu Thr Ala Ser Gly Thr Phe Gln  
 20 25 30  
 Asp Asp Asp Ile Lys Leu Asn His Thr Gly Leu Arg Val Val Ser Ser  
 35 40 45  
 Glu Pro Asn Leu Pro Thr Gln Thr Gln Ser Ser Ser Pro Asp Gly Gln  
 50 55 60  
 Leu Ser Ile Ala Asp Leu Glu Leu Val Arg Phe Leu Gly Lys Gly Ala  
 65 70 75 80  
 Gly Gly Thr Val Gln Leu Val Arg His Lys Trp Thr Asn Val Asn Tyr  
 85 90 95  
 Ala Leu Lys Ala Ile Gln Met Asn Ile Asn Glu Thr Val Arg Lys Gln  
 100 105 110

-continued

Ile Val Gln Glu Leu Lys Ile Asn Gln Val Thr His Gln Gln Cys Pro  
 115 120 125  
 Tyr Ile Val Glu Cys Phe His Ser Phe Tyr His Asn Gly Val Ile Ser  
 130 135 140  
 Met Ile Leu Glu Tyr Met Asp Arg Gly Ser Leu Ser Asp Ile Ile Lys  
 145 150 155 160  
 Gln Gln Lys Gln Ile Pro Glu Pro Tyr Leu Ala Val Ile Ala Ser Gln  
 165 170 175  
 Val Leu Lys Gly Leu Glu Tyr Leu His Gln Val Arg His Ile Ile His  
 180 185 190  
 Arg Asp Ile Lys Pro Ser Asn Leu Leu Ile Asn His Lys Gly Glu Val  
 195 200 205  
 Lys Ile Ser Asp Phe Gly Val Ser Ala Val Leu Val His Ser Leu Ala  
 210 215 220  
 Gln Arg Asp Thr Phe Val Gly Thr Cys Thr Tyr Met Ser Pro Glu Arg  
 225 230 235 240  
 Leu Gln Gly Arg Ser Tyr Ala Tyr Asp Ser Asp Leu Trp Ser Leu Gly  
 245 250 255  
 Leu Thr Leu Leu Glu Cys Ala Leu Gly Thr Phe Pro Tyr Lys Pro Ala  
 260 265 270  
 Gly Met Glu Glu Gly Trp Gln Asn Phe Phe Ile Leu Met Glu Cys Ile  
 275 280 285  
 Val Asn Gln Pro Pro Ala Ala Ala Ser Pro Asp Lys Phe Ser Pro Glu  
 290 295 300  
 Phe Cys Ser Phe Ile Glu Ser Cys Ile Arg Lys Cys Pro Ser Glu Arg  
 305 310 315 320  
 Pro Ser Thr Thr Asp Leu Leu Lys His Pro Phe Leu Gln Lys Tyr Asn  
 325 330 335  
 Glu Glu Glu Tyr His Leu Ser Lys Ile Leu  
 340 345

&lt;210&gt; SEQ ID NO 37

&lt;211&gt; LENGTH: 346

&lt;212&gt; TYPE: PRT

&lt;213&gt; ORGANISM: Physcomitrella patens

&lt;400&gt; SEQUENCE: 37

Met Ser Arg Arg Val Arg Arg Gly Gly Leu Arg Val Ala Val Pro Lys  
 1 5 10 15  
 Gln Glu Thr Pro Val Ser Lys Phe Leu Thr Ala Ser Gly Thr Phe Gln  
 20 25 30  
 Asp Asp Asp Ile Lys Leu Asn His Thr Gly Leu Arg Val Val Ser Ser  
 35 40 45  
 Glu Pro Asn Leu Pro Thr Gln Thr Gln Ser Ser Ser Pro Asp Gly Gln  
 50 55 60  
 Leu Ser Ile Ala Asp Leu Glu Leu Val Arg Phe Leu Gly Lys Gly Ala  
 65 70 75 80  
 Gly Gly Thr Val Gln Leu Val Arg His Lys Trp Thr Asn Val Asn Tyr  
 85 90 95  
 Ala Leu Lys Ala Ile Gln Met Asn Ile Asn Glu Thr Val Arg Lys Gln  
 100 105 110  
 Ile Val Gln Glu Leu Lys Ile Asn Gln Val Thr His Gln Gln Cys Pro  
 115 120 125  
 Tyr Ile Val Glu Cys Phe His Ser Phe Tyr His Asn Gly Val Ile Ser  
 130 135 140

-continued

Met Ile Leu Glu Tyr Met Asp Arg Gly Ser Leu Ser Asp Ile Ile Lys  
 145 150 155 160  
 Gln Gln Lys Gln Ile Pro Glu Pro Tyr Leu Ala Val Ile Ala Ser Gln  
 165 170 175  
 Val Leu Lys Gly Leu Glu Tyr Leu His Gln Val Arg His Ile Ile His  
 180 185 190  
 Arg Asp Ile Lys Pro Ser Asn Leu Leu Ile Asn His Lys Gly Glu Val  
 195 200 205  
 Lys Ile Ser Asp Phe Gly Val Ser Ala Val Leu Val His Ser Leu Ala  
 210 215 220  
 Gln Arg Asp Thr Phe Val Gly Thr Cys Thr Tyr Met Ser Pro Glu Arg  
 225 230 235 240  
 Leu Gln Gly Arg Ser Tyr Ala Tyr Asp Ser Asp Leu Trp Ser Leu Gly  
 245 250 255  
 Leu Thr Leu Leu Glu Cys Ala Leu Gly Thr Phe Pro Tyr Lys Pro Ala  
 260 265 270  
 Gly Met Glu Glu Gly Trp Gln Asn Phe Phe Ile Leu Met Glu Cys Ile  
 275 280 285  
 Val Asn Gln Pro Pro Ala Ala Ala Ser Pro Asp Lys Phe Ser Pro Glu  
 290 295 300  
 Phe Cys Ser Phe Ile Glu Ser Cys Ile Arg Lys Cys Pro Ser Glu Arg  
 305 310 315 320  
 Pro Ser Thr Thr Asp Leu Leu Lys His Pro Phe Leu Gln Lys Tyr Asn  
 325 330 335  
 Glu Glu Glu Tyr His Leu Ser Lys Ile Leu  
 340 345

&lt;210&gt; SEQ ID NO 38

&lt;211&gt; LENGTH: 597

&lt;212&gt; TYPE: PRT

&lt;213&gt; ORGANISM: Physcomitrella patens

&lt;400&gt; SEQUENCE: 38

Met Gly Gln Cys Tyr Gly Lys Phe Asp Asp Gly Gly Glu Gly Glu Asp  
 1 5 10 15  
 Leu Phe Glu Arg Gln Lys Val Gln Val Ser Arg Thr Pro Lys His Gly  
 20 25 30  
 Ser Trp Ser Asn Ser Asn Arg Gly Ser Phe Asn Asn Gly Gly Gly Ala  
 35 40 45  
 Ser Pro Met Arg Ala Lys Thr Ser Phe Gly Ser Ser His Pro Ser Pro  
 50 55 60  
 Arg His Pro Ser Ala Ser Pro Leu Pro His Tyr Thr Ser Ser Pro Ala  
 65 70 75 80  
 Pro Ser Thr Pro Arg Arg Asn Ile Phe Lys Arg Pro Phe Pro Pro Pro  
 85 90 95  
 Ser Pro Ala Lys His Ile Gln Ser Ser Leu Val Lys Arg His Gly Ala  
 100 105 110  
 Lys Pro Lys Glu Gly Gly Ala Ile Pro Glu Ala Val Asp Gly Glu Lys  
 115 120 125  
 Pro Leu Asp Lys His Phe Gly Tyr His Lys Asn Phe Ala Thr Lys Tyr  
 130 135 140  
 Glu Leu Gly His Glu Val Gly Arg Gly His Phe Gly His Thr Cys Tyr  
 145 150 155 160  
 Ala Lys Val Arg Lys Gly Glu His Lys Gly Gln Ala Val Ala Val Lys  
 165 170 175

-continued

---

Ile Ile Ser Lys Ala Lys Met Thr Thr Ala Ile Ala Ile Glu Asp Val  
                   180  185  190

Gly Arg Glu Val Lys Ile Leu Lys Ala Leu Thr Gly His Gln Asn Leu  
                   195  200  205

Val Arg Phe Tyr Asp Ser Cys Glu Asp His Leu Asn Val Tyr Ile Val  
                   210  215  220

Met Glu Leu Cys Glu Gly Gly Glu Leu Leu Asp Arg Ile Leu Ser Arg  
                   225  230  235  240

Gly Gly Lys Tyr Ser Glu Glu Asp Ala Lys Val Val Val Arg Gln Ile  
                                   245  250  255

Leu Ser Val Val Ala Phe Cys His Leu Gln Gly Val Val His Arg Asp  
                   260  265  270

Leu Lys Pro Glu Asn Phe Leu Phe Thr Thr Lys Asp Glu Tyr Ala Gln  
                   275  280  285

Leu Lys Ala Ile Asp Phe Gly Leu Ser Asp Phe Ile Lys Pro Asp Glu  
                   290  295  300

Arg Leu Asn Asp Ile Val Gly Ser Ala Tyr Tyr Val Ala Pro Glu Val  
                   305  310  315  320

Leu His Arg Leu Tyr Ser Met Glu Ala Asp Val Trp Ser Ile Gly Val  
                                   325  330  335

Ile Thr Tyr Ile Leu Leu Cys Gly Ser Arg Pro Phe Trp Ala Arg Thr  
                   340  345  350

Glu Ser Gly Ile Phe Arg Ala Val Leu Arg Ala Asp Pro Ser Phe Glu  
                   355  360  365

Glu Ala Pro Trp Pro Ser Ile Ser Pro Glu Ala Lys Asp Phe Val Lys  
                   370  375  380

Arg Leu Leu Asn Lys Asp Met Arg Lys Arg Met Thr Ala Ala Gln Ala  
                   385  390  395  400

Leu Thr His Pro Trp Ile Arg Ser Asn Asn Val Lys Ile Pro Leu Asp  
                                   405  410  415

Ile Leu Val Tyr Arg Leu Val Arg Asn Tyr Leu Arg Ala Ser Ser Met  
                   420  425  430

Arg Lys Ala Ala Leu Lys Ala Leu Ser Lys Thr Leu Thr Glu Asp Glu  
                   435  440  445

Thr Phe Tyr Leu Arg Thr Gln Phe Met Leu Leu Glu Pro Ser Asn Asn  
                   450  455  460

Gly Arg Val Thr Phe Glu Asn Phe Arg Gln Ala Leu Leu Lys Asn Ser  
                   465  470  475  480

Thr Glu Ala Met Lys Glu Ser Arg Val Phe Glu Ile Leu Glu Ser Met  
                                   485  490  495

Asp Gly Leu His Phe Lys Lys Met Asp Phe Ser Glu Phe Cys Ala Ala  
                                   500  505  510

Ala Ile Ser Val Leu Gln Leu Glu Ala Thr Glu Arg Trp Glu Gln His  
                   515  520  525

Ala Arg Ala Ala Tyr Asp Ile Phe Glu Lys Glu Gly Asn Arg Val Ile  
                   530  535  540

Tyr Pro Asp Glu Leu Ala Lys Glu Met Gly Leu Ala Pro Asn Val Pro  
                   545  550  555  560

Ala Gln Val Phe Leu Asp Trp Ile Arg Gln Ser Asp Gly Arg Leu Ser  
                                   565  570  575

Phe Thr Gly Phe Thr Lys Leu Leu His Gly Ile Ser Ser Arg Ala Ile  
                   580  585  590

Lys Asn Leu Gln Gln

-continued

595

```

<210> SEQ ID NO 39
<211> LENGTH: 549
<212> TYPE: PRT
<213> ORGANISM: Physcomitrella patens

<400> SEQUENCE: 39
Met Gly Asn Thr Ser Ser Arg Gly Ser Arg Lys Ser Thr Arg Gln Val
 1          5          10          15
Asn Gln Gly Val Gly Ser Gln Asp Thr Arg Glu Lys Asn Asp Ser Val
 20          25          30
Asn Pro Lys Thr Arg Gln Gly Gly Ser Val Gly Ala Asn Asn Tyr Gly
 35          40          45
Gly Lys Pro Ser Ser Gly Ala Gln Ala Gly Glu Arg Ser Thr Ser Ala
 50          55          60
Pro Ala Ala Leu Pro Arg Pro Lys Pro Ala Ser Arg Ser Val Ser Gly
 65          70          75          80
Val Leu Gly Lys Pro Leu Ser Asp Ile Arg Gln Ser Tyr Ile Leu Gly
 85          90          95
Arg Glu Leu Gly Arg Gly Gln Phe Gly Val Thr Tyr Leu Cys Thr Asp
 100         105         110
Lys Met Thr Asn Glu Ala Tyr Ala Cys Lys Ser Ile Ala Lys Arg Lys
 115         120         125
Leu Thr Ser Lys Glu Asp Ile Glu Asp Val Lys Arg Glu Val Gln Ile
 130         135         140
Met His His Leu Ser Gly Thr Pro Asn Ile Val Val Leu Lys Asp Val
 145         150         155         160
Phe Glu Asp Lys His Ser Val His Leu Val Met Glu Leu Cys Ala Gly
 165         170         175
Gly Glu Leu Phe Asp Arg Ile Ile Ala Lys Gly His Tyr Ser Glu Arg
 180         185         190
Ala Ala Ala Asp Met Cys Arg Val Ile Val Asn Val Val His Arg Cys
 195         200         205
His Ser Leu Gly Val Phe His Arg Asp Leu Lys Pro Glu Asn Phe Leu
 210         215         220
Leu Ala Ser Lys Ala Glu Asp Ala Pro Leu Lys Ala Thr Asp Phe Gly
 225         230         235         240
Leu Ser Thr Phe Phe Lys Pro Gly Asp Val Phe Gln Asp Ile Val Gly
 245         250         255
Ser Ala Tyr Tyr Val Ala Pro Glu Val Leu Lys Arg Ser Tyr Gly Pro
 260         265         270
Glu Ala Asp Val Trp Ser Ala Gly Val Ile Val Tyr Ile Leu Leu Cys
 275         280         285
Gly Val Pro Pro Phe Trp Ala Glu Thr Glu Gln Gly Ile Phe Asp Ala
 290         295         300
Val Leu Lys Gly His Ile Asp Phe Glu Asn Asp Pro Trp Pro Lys Ile
 305         310         315         320
Ser Asn Gly Ala Lys Asp Leu Val Arg Lys Met Leu Asn Pro Asn Val
 325         330         335
Lys Ile Arg Leu Thr Ala Gln Gln Val Leu Asn His Pro Trp Met Lys
 340         345         350
Glu Asp Gly Asp Ala Pro Asp Val Pro Leu Asp Asn Ala Val Leu Thr
 355         360         365
Arg Leu Lys Asn Phe Ser Ala Ala Asn Lys Met Lys Lys Leu Ala Leu

```

-continued

370	375	380
Lys Val Ile Ala Glu Ser Leu Ser Glu Glu Glu Ile Val Gly Leu Arg 385 390 395 400		
Glu Met Phe Lys Ser Ile Asp Thr Asp Asn Ser Gly Thr Val Thr Phe 405 410 415		
Glu Glu Leu Lys Glu Gly Leu Leu Lys Gln Gly Ser Lys Leu Asn Glu 420 425 430		
Ser Asp Ile Arg Lys Leu Met Glu Ala Ala Asp Val Asp Gly Asn Gly 435 440 445		
Lys Ile Asp Phe Asn Glu Phe Ile Ser Ala Thr Met His Met Asn Lys 450 455 460		
Thr Glu Lys Glu Asp His Leu Trp Ala Ala Phe Met His Phe Asp Thr 465 470 475 480		
Asp Asn Ser Gly Tyr Ile Thr Ile Asp Glu Leu Gln Glu Ala Met Glu 485 490 495		
Lys Asn Gly Met Gly Asp Pro Glu Thr Ile Gln Glu Ile Ile Ser Glu 500 505 510		
Val Asp Thr Asp Asn Asp Gly Arg Ile Asp Tyr Asp Glu Phe Val Ala 515 520 525		
Met Met Arg Lys Gly Asn Pro Gly Ala Glu Asn Gly Gly Thr Val Asn 530 535 540		
Lys Pro Arg His Arg 545		

<210> SEQ ID NO 40  
 <211> LENGTH: 18  
 <212> TYPE: DNA  
 <213> ORGANISM: Artificial Sequence  
 <220> FEATURE:  
 <223> OTHER INFORMATION: Description of Artificial Sequence: Primer

<400> SEQUENCE: 40

caggaaacag ctatgacc

18

<210> SEQ ID NO 41  
 <211> LENGTH: 19  
 <212> TYPE: DNA  
 <213> ORGANISM: Artificial Sequence  
 <220> FEATURE:  
 <223> OTHER INFORMATION: Description of Artificial Sequence: Primer

<400> SEQUENCE: 41

ctaaagggaa caaaagctg

19

<210> SEQ ID NO 42  
 <211> LENGTH: 18  
 <212> TYPE: DNA  
 <213> ORGANISM: Artificial Sequence  
 <220> FEATURE:  
 <223> OTHER INFORMATION: Description of Artificial Sequence: Primer

<400> SEQUENCE: 42

tgtaaaacga cggccagt

18

<210> SEQ ID NO 43  
 <211> LENGTH: 25  
 <212> TYPE: DNA  
 <213> ORGANISM: Artificial Sequence  
 <220> FEATURE:  
 <223> OTHER INFORMATION: Description of Artificial Sequence: Primer

<400> SEQUENCE: 43



-continued

---

ccacggctctt cggtgctgg tcgtg 25

<210> SEQ ID NO 44  
<211> LENGTH: 25  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer

<400> SEQUENCE: 44

gcagcacagc accaccagcg gctat 25

<210> SEQ ID NO 45  
<211> LENGTH: 25  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer

<400> SEQUENCE: 45

gcgcccagtg agtagctcca gcatt 25

<210> SEQ ID NO 46  
<211> LENGTH: 30  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer

<400> SEQUENCE: 46

atcccgggtg agtatcactt acggtggcga 30

<210> SEQ ID NO 47  
<211> LENGTH: 33  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer

<400> SEQUENCE: 47

gcgttaactc gaccaaggtc actattccaa gca 33

<210> SEQ ID NO 48  
<211> LENGTH: 25  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer

<400> SEQUENCE: 48

cggtgcccac ctggttctg tggtt 25

<210> SEQ ID NO 49  
<211> LENGTH: 31  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer

<400> SEQUENCE: 49

atcccgggag tgggtggttg gactgtaagg a 31

<210> SEQ ID NO 50  
<211> LENGTH: 34  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence

-continued

---

<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
<400> SEQUENCE: 50  
gcgtaaacct tcgtcttgga caggtagagg ttac 34

<210> SEQ ID NO 51  
<211> LENGTH: 25  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
<400> SEQUENCE: 51  
gactcagccc cgtaatcctt caaca 25

<210> SEQ ID NO 52  
<211> LENGTH: 31  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
<400> SEQUENCE: 52  
atcccgggca acgagaagca ttcgagatgg c 31

<210> SEQ ID NO 53  
<211> LENGTH: 33  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
<400> SEQUENCE: 53  
gcgtaaacga gcatcacgat actcggatg ttc 33

<210> SEQ ID NO 54  
<211> LENGTH: 27  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
<400> SEQUENCE: 54  
cgacggctaa taccacgttg gcgacca 27

<210> SEQ ID NO 55  
<211> LENGTH: 33  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
<400> SEQUENCE: 55  
atcccgggct gtgatgtcgg tgtgggtgctc tgc 33

<210> SEQ ID NO 56  
<211> LENGTH: 34  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
<400> SEQUENCE: 56  
gcgagctcgc accactgaat gatggagact cagg 34

-continued

---

<210> SEQ ID NO 57  
<211> LENGTH: 25  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 57  
  
cgaccgcagc ccatgaggaa gttat 25

<210> SEQ ID NO 58  
<211> LENGTH: 33  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 58  
  
atccccgggct cacgtagtgc actgaactct gtc 33

<210> SEQ ID NO 59  
<211> LENGTH: 33  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 59  
  
gcgttaacat gcccatcttc tcatactcag acc 33

<210> SEQ ID NO 60  
<211> LENGTH: 25  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 60  
  
ctcgcctacc aagccccatt agaaa 25

<210> SEQ ID NO 61  
<211> LENGTH: 32  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 61  
  
atccccgggtt gtcgaggacg gagagagaag ag 32

<210> SEQ ID NO 62  
<211> LENGTH: 33  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 62  
  
gcgttaacct taggaatcgt atggcagaga gct 33

<210> SEQ ID NO 63  
<211> LENGTH: 25  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 63

-continued

---

gcttcacaat gttgggcct ccaca 25

<210> SEQ ID NO 64  
<211> LENGTH: 33  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer

<400> SEQUENCE: 64

gcgtaacgg gaggaagtc ggggaagag acg 33

<210> SEQ ID NO 65  
<211> LENGTH: 33  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer

<400> SEQUENCE: 65

gcgagctcag cgcttcgac aactgagaaa cct 33

<210> SEQ ID NO 66  
<211> LENGTH: 25  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer

<400> SEQUENCE: 66

acgagaaggt tgggggctt caagt 25

<210> SEQ ID NO 67  
<211> LENGTH: 30  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer

<400> SEQUENCE: 67

atcccggcg agccatggcg ccacttgctt 30

<210> SEQ ID NO 68  
<211> LENGTH: 33  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer

<400> SEQUENCE: 68

gcgtaacgc cgagcaaaa tgtctgctgg atg 33

<210> SEQ ID NO 69  
<211> LENGTH: 25  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer

<400> SEQUENCE: 69

cccggtaagc catcggagtg tggaa 25

<210> SEQ ID NO 70  
<211> LENGTH: 30  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence

-continued

---

<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
<400> SEQUENCE: 70  
atcccgggct tgtattggct cggataattt 30

<210> SEQ ID NO 71  
<211> LENGTH: 33  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
<400> SEQUENCE: 71  
gcgtaaacgg caatatctgc acagccgttc act 33

<210> SEQ ID NO 72  
<211> LENGTH: 25  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
<400> SEQUENCE: 72  
gtgtctcgcgt gggccaagga atgaa 25

<210> SEQ ID NO 73  
<211> LENGTH: 35  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
<400> SEQUENCE: 73  
atcccgggcg gtcgagtcgt attaggtgtt gtttc 35

<210> SEQ ID NO 74  
<211> LENGTH: 30  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
<400> SEQUENCE: 74  
gagctccggt aggtccgacc ttttcaattg 30

<210> SEQ ID NO 75  
<211> LENGTH: 26  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
<400> SEQUENCE: 75  
gacgacgcga agcccgggtg ggttga 26

<210> SEQ ID NO 76  
<211> LENGTH: 31  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
<400> SEQUENCE: 76  
atcccgggag aggtgatct gatgctacag t 31

-continued

---

<210> SEQ ID NO 77  
 <211> LENGTH: 33  
 <212> TYPE: DNA  
 <213> ORGANISM: Artificial Sequence  
 <220> FEATURE:  
 <223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
 <400> SEQUENCE: 77  
  
 atgagctctg gcggattggc gaggtagttc gac 33

<210> SEQ ID NO 78  
 <211> LENGTH: 25  
 <212> TYPE: DNA  
 <213> ORGANISM: Artificial Sequence  
 <220> FEATURE:  
 <223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
 <400> SEQUENCE: 78  
  
 cggcgcaacy tagtatgcgc ttcca 25

<210> SEQ ID NO 79  
 <211> LENGTH: 27  
 <212> TYPE: DNA  
 <213> ORGANISM: Artificial Sequence  
 <220> FEATURE:  
 <223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
 <400> SEQUENCE: 79  
  
 cgcggtgaac aacaccttgc aggtgac 27

<210> SEQ ID NO 80  
 <211> LENGTH: 25  
 <212> TYPE: DNA  
 <213> ORGANISM: Artificial Sequence  
 <220> FEATURE:  
 <223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
 <400> SEQUENCE: 80  
  
 gctcgggtca gcctcaaca ccgca 25

<210> SEQ ID NO 81  
 <211> LENGTH: 25  
 <212> TYPE: DNA  
 <213> ORGANISM: Artificial Sequence  
 <220> FEATURE:  
 <223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
 <400> SEQUENCE: 81  
  
 gttaaagctt gtgcagcagt catgc 25

<210> SEQ ID NO 82  
 <211> LENGTH: 31  
 <212> TYPE: DNA  
 <213> ORGANISM: Artificial Sequence  
 <220> FEATURE:  
 <223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
 <400> SEQUENCE: 82  
  
 atcccgggtg tagggggcg aggttcgatg c 31

<210> SEQ ID NO 83  
 <211> LENGTH: 34  
 <212> TYPE: DNA  
 <213> ORGANISM: Artificial Sequence  
 <220> FEATURE:  
 <223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
 <400> SEQUENCE: 83

-continued

---

gcgттаacga caaccggagt agaacggcag тса 34

<210> SEQ ID NO 84  
<211> LENGTH: 25  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer

<400> SEQUENCE: 84

agaagcgagg aatgggcagg gacga 25

<210> SEQ ID NO 85  
<211> LENGTH: 32  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer

<400> SEQUENCE: 85

atcccgggcg aactgcgatc tgagattcca ac 32

<210> SEQ ID NO 86  
<211> LENGTH: 34  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer

<400> SEQUENCE: 86

gcgттаacga gatccaaccg aagccatcct acga 34

<210> SEQ ID NO 87  
<211> LENGTH: 30  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer

<400> SEQUENCE: 87

gcgctgcaga tttcatttgg agaggacacg 30

<210> SEQ ID NO 88  
<211> LENGTH: 35  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer

<400> SEQUENCE: 88

cgcgcccggc ctcagaagaa ctcgtcaaga aggcg 35

<210> SEQ ID NO 89  
<211> LENGTH: 25  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer

<400> SEQUENCE: 89

gctgacacgc caagcctcgc tagtc 25

<210> SEQ ID NO 90  
<211> LENGTH: 33  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence

-continued

---

<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
<400> SEQUENCE: 90  
gcgtaactc gaccaaggtc actattccaa gca 33

<210> SEQ ID NO 91  
<211> LENGTH: 34  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
<400> SEQUENCE: 91  
gcgtaacct tcgtcttggc caggtagg ttac 34

<210> SEQ ID NO 92  
<211> LENGTH: 33  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
<400> SEQUENCE: 92  
gcgtaacga gcatcagat actcgggat ttc 33

<210> SEQ ID NO 93  
<211> LENGTH: 34  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
<400> SEQUENCE: 93  
gcgagctgc accactgaat gatggagact cagg 34

<210> SEQ ID NO 94  
<211> LENGTH: 33  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
<400> SEQUENCE: 94  
gcgtaacat gccatcttc tcatactcag acc 33

<210> SEQ ID NO 95  
<211> LENGTH: 33  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
<400> SEQUENCE: 95  
gcgtaacct taggaatcgt atggcagaga gct 33

<210> SEQ ID NO 96  
<211> LENGTH: 33  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
<400> SEQUENCE: 96  
gcgagctcag cgcttcgcac aactgagaaa cct 33



-continued

---

<210> SEQ ID NO 97  
<211> LENGTH: 33  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 97  
  
gcgттаacgg caatatctgc acagcggttc act 33

<210> SEQ ID NO 98  
<211> LENGTH: 33  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 98  
  
gcgттаacgg caatatctgc acagcggttc act 33

<210> SEQ ID NO 99  
<211> LENGTH: 30  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 99  
  
gagctccggt aggtccgacc tcttcaattg 30

<210> SEQ ID NO 100  
<211> LENGTH: 33  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 100  
  
atgagctctg gcgгattggc gaggtagttc gac 33

<210> SEQ ID NO 101  
<211> LENGTH: 34  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 101  
  
gcgттаacga caaccggagt agaacggcag тсса 34

<210> SEQ ID NO 102  
<211> LENGTH: 34  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 102  
  
gcgттаacga gatccaaccg aagccatcct acga 34

<210> SEQ ID NO 103  
<211> LENGTH: 25  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 103

-continued

---

cccagtaata gcagggttg aggaa 25

<210> SEQ ID NO 104  
<211> LENGTH: 25  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer

<400> SEQUENCE: 104

ggctgctga agatccgcta cagag 25

<210> SEQ ID NO 105  
<211> LENGTH: 25  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer

<400> SEQUENCE: 105

cgtcaggcta ctttgctgg agcac 25

<210> SEQ ID NO 106  
<211> LENGTH: 25  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer

<400> SEQUENCE: 106

cggtgctggc taacaccagg ccaga 25

<210> SEQ ID NO 107  
<211> LENGTH: 31  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer

<400> SEQUENCE: 107

atcccgggca acgagaagca ttcgagatgg c 31

<210> SEQ ID NO 108  
<211> LENGTH: 33  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer

<400> SEQUENCE: 108

gcgttaacga gcatcacgat actcggatgat ttc 33

<210> SEQ ID NO 109  
<211> LENGTH: 25  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer

<400> SEQUENCE: 109

cgtggcatct ctcccgatgt tctta 25

<210> SEQ ID NO 110  
<211> LENGTH: 25  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence

-continued

---

<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 110  
  
ggccaactga aggcgtgtca tgatc 25

<210> SEQ ID NO 111  
<211> LENGTH: 25  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 111  
  
ctcgaggget cgttcaccgt gacct 25

<210> SEQ ID NO 112  
<211> LENGTH: 26  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 112  
  
cggaggtaac agtagtcagg ctgctc 26

<210> SEQ ID NO 113  
<211> LENGTH: 25  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 113  
  
ccgcgaccct tccacgcac agcat 25

<210> SEQ ID NO 114  
<211> LENGTH: 25  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 114  
  
cctccaggaa gctgcgccg agaag 25

<210> SEQ ID NO 115  
<211> LENGTH: 26  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 115  
  
ggacattgtc cgtgatcagc aatcga 26

<210> SEQ ID NO 116  
<211> LENGTH: 25  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 116  
  
cagcctctgg aacaaccaga cgctg 25

-continued

---

<210> SEQ ID NO 117  
<211> LENGTH: 25  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 117  
  
gtcaccgcga ggtacaagcc accac 25

<210> SEQ ID NO 118  
<211> LENGTH: 25  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 118  
  
gcagctctgg agctctgtac cacct 25

<210> SEQ ID NO 119  
<211> LENGTH: 25  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 119  
  
acggccacgt cgagaatctg agcaa 25

<210> SEQ ID NO 120  
<211> LENGTH: 25  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 120  
  
cgaagtgctc gcaagcaatg ccgaa 25

<210> SEQ ID NO 121  
<211> LENGTH: 35  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 121  
  
atcccgggcg gtcgagtcgt attagtggtt gtttc 35

<210> SEQ ID NO 122  
<211> LENGTH: 30  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 122  
  
gagctccggt aggtccgacc tcttcaattg 30

<210> SEQ ID NO 123  
<211> LENGTH: 26

-continued

---

<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 123  
  
gggcaactgt caatagcaga cctgga 26

<210> SEQ ID NO 124  
<211> LENGTH: 26  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 124  
  
gcaagtccca acgaacgtgt ctcgct 26

<210> SEQ ID NO 125  
<211> LENGTH: 25  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 125  
  
gcgaagatga cgactgctat tgcca 25

<210> SEQ ID NO 126  
<211> LENGTH: 25  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 126  
  
cgtgatgact ccaatgctcc atacg 25

<210> SEQ ID NO 127  
<211> LENGTH: 27  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 127  
  
gccagcateg aggtcagtat ccggtgt 27

<210> SEQ ID NO 128  
<211> LENGTH: 27  
<212> TYPE: DNA  
<213> ORGANISM: Artificial Sequence  
<220> FEATURE:  
<223> OTHER INFORMATION: Description of Artificial Sequence: Primer  
  
<400> SEQUENCE: 128  
  
gtctgtggcc ttcagagcgc catcctc 27

---

## 181

The invention claimed is:

1. A transgenic plant cell transformed with an expression vector comprising an isolated polynucleotide selected from the group consisting of:

a) a polynucleotide having a sequence comprising nucleotides 1 to 2784 of SEQ ID NO:14; and

b) a polynucleotide encoding a polypeptide having a sequence comprising amino acids 1 to 749 of SEQ ID NO:27.

2. The plant cell of claim 1, wherein the polynucleotide has the sequence comprising nucleotides 1 to 2784 of SEQ ID NO:14.

3. The plant cell of claim 1, wherein the polynucleotide encodes the polypeptide having the sequence comprising amino acids 1 to 749 of SEQ ID NO:27.

4. A transgenic plant transformed with an expression cassette comprising an isolated polynucleotide selected from the group consisting of:

a) a polynucleotide having a sequence comprising nucleotides 1 to 2784 of SEQ ID NO:14; and

b) a polynucleotide encoding a polypeptide having a sequence comprising amino acids 1 to 749 of SEQ ID NO:27.

5. The plant of claim 4, wherein the polynucleotide has the sequence comprising nucleotides 1 to 2784 of SEQ ID NO:14.

6. The plant of claim 4, wherein the polynucleotide encodes the polypeptide having the sequence comprising amino acids 1 to 749 of SEQ ID NO:27.

7. The plant of claim 4, further described as a monocot.

8. The plant of claim 5, further described as a dicot.

9. The plant of claim 4, wherein the plant is selected from the group consisting of maize, wheat, rye, oat, triticale, rice, barley, soybean, peanut, cotton, rapeseed, canola, manihot, pepper, sunflower, tagetes, potato, tobacco, eggplant, tomato, *Vicia* species, pea, alfalfa, coffee, cacao, tea, *Salix* species, oil palm, coconut, perennial grasses, and a forage crop plant.

10. The plant of claim 9, which is maize.

11. The plant of claim 9, which is soybean.

12. The plant of claim 9, which is rapeseed or canola.

13. The plant of claim 9, which is cotton.

14. A seed which is true breeding for a transgene comprising a polynucleotide selected from the group consisting of:

## 182

a) a polynucleotide having a sequence comprising nucleotides 1 to 2784 of SEQ ID NO:14; and

b) a polynucleotide encoding a polypeptide having a sequence comprising amino acids 1 to 749 of SEQ ID NO:27.

15. The seed of claim 14, wherein the polynucleotide has the sequence comprising nucleotides 1 to 2784 of SEQ ID NO:14.

16. The seed of claim 14, wherein the polynucleotide encodes the polypeptide having the sequence comprising amino acids 1 to 749 of SEQ ID NO:27.

17. An isolated nucleic acid comprising a polynucleotide selected from the group consisting of:

a) a polynucleotide having a sequence comprising nucleotides 1 to 2784 of SEQ ID NO:14; and

b) a polynucleotide encoding a polypeptide having a sequence comprising amino acids 1 to 749 of SEQ ID NO:27.

18. The isolated nucleic acid of claim 17, wherein the polynucleotide has the sequence comprising nucleotides 1 to 2784 of SEQ ID NO:14.

19. The isolated nucleic acid of claim 17, wherein the polynucleotide encodes the polypeptide having the sequence comprising amino acids 1 to 749 of SEQ ID NO:27.

20. A method of producing a drought-tolerant transgenic plant, the method comprising the steps of:

a) transforming a plant cell with an expression vector comprising a polynucleotide selected from the group consisting of:

a) a polynucleotide having a sequence comprising nucleotides 1 to 2784 of SEQ ID NO:14; and

b) a polynucleotide encoding a polypeptide having a sequence comprising amino acids 1 to 749 of SEQ ID NO:27.

21. The method of claim 20, wherein the polynucleotide has the sequence comprising nucleotides 1 to 2784 of SEQ ID NO:14.

22. The method of claim 20, wherein the polynucleotide encodes the polypeptide having the sequence comprising amino acids 1 to 749 of SEQ ID NO:27.

\* \* \* \* \*